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Investigation

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# BUNCHY TOP DISEASE

OF THE

# **BANANA**

By C. J. P. MAGEE, B.Sc.Agr.

MELBOURNE, 1927

By Authority:

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### FOREWORD.

This Bulletin deals with the scientific aspect of the Bunchy Top Disease in bananas, and is the result of an investigation instituted by the Governments of the Commonwealth, New South Wales, and Queensland. I have considered it fitting and proper that this Bulletin should be issued as the work of the Assistant Plant Pathologist, Mr. C. J. P. Magee, B.Sc. Agr., since the knowledge of the nature and mode of transmission of the disease is mainly the result of investigations carried out by that officer. A general report dealing with the problem has already been issued. Further investigations regarding certain aspects of the problem are still in progress.

E. J. GODDARD, Supervisor.

Department of Biology, University of Queensland, Brisbane.



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## Investigations on the Bunchy Top Disease of the Banana.

### 1. INTRODUCTION.

It is the purpose of this paper to record the results obtained during the past eighteen months in the investigation of the Bunchy Top disease in the banana. This disease was first reported in Australia about 1913, and has since practically wiped out the banana industry in many centres in north-eastern New South Wales and south-eastern Queensland. It is doubtful if there are many other diseases on record which have wrought such devastation on a flourishing industry as that produced by the ravages of Bunchy Top. The disease has been easily the most serious banana malady in Australia, and has also occasioned serious losses in Fiji, Ceylon, and Egypt.

Up till the time of the present investigation, nothing definite was known regarding the nature of the cause of this disease or of its means of natural transmission. The absence of this knowledge has been the indirect cause of the rapid extension of the disease and its calamitous effects over such a wide area. Also, until the present time, it has not been possible to elaborate any reliable scheme of control measures to check the ravages of the disease. Evidence is contained in this paper which definitely proves that Bunchy Top is an insect-transmitted disease belonging to the class of virus diseases.

The Bunchy Top Investigation, consisting of Professor E. J. Goddard, B.A., D.Sc. (Supervisor), the writer (Assistant Plant Pathologist), and Mr. H. Collard (Horticulturist), undertook the responsibility of researches on Bunchy Top in May, 1924, on the recommendation of a Board which represented the Commonwealth Institute of Science and Industry, the New South Wales Department of Agriculture, and the Queensland Department of Agriculture and Stock. The funds required for the investigations were contributed by these three bodies, and the expenditure of the money was controlled by the Board.

A considerable amount of work had previously been devoted to the problem by scientific workers in Australia, as well as in Fiji (where the disease has been present for at least 40 years), Ceylon, Egypt, and the

The Supervisor of the Bunchy Top Investigation, Professor E. J. Goddard, has graciously suggested that I should accept the responsibility of preparing this paper. In doing so I feel that I am under a great obligation to him and to the Hortieulturist, Mr. H. Collard. To Profess Goddard I am indebted for help in every direction during the course of the Investigation and for criticism, suggestions and assistance in the preparation of the manuscript. To Mr. Collard I owe a great deal for his care and thoroughness in field work, for observations and practical suggestions, and constant co-operation during the work. I desire to make acknowledgment of the kind assistance received from Mr. W. L. Waterhouse, University of Sydney, by the loan of literature and for helpful suggestions throughout the Investigation. Thanks are also due to Mr. A. E. Mee for assistance in photography.

—C. J. P. Magee.

Philippines, but no definite cause could be proved. It had been suggested by some that the disease was caused by nematodes (in Fiji, Egypt, and the Philippines), while various others had suggested that the causal agent was a fungus, chemical deficiency of the soil, deterioration of the banana stock, climatic factors, aphides, &c. There was no substantive evidence to strengthen the claims of these opinions, and as the ravages of the disease along the northern rivers of New South Wales, and in south-eastern Queensland, became so intense, it was clear that any attempt to solve the problem would have to be made by scientific investigators working on the spot.

A laboratory was founded at Tweed Heads, and experimental plots were leased at Cobaki from Messrs. T. Pilgrim and McAlister. The investigation work at the laboratory was attended to by the writer, and the horticultural work at the plots was under the care of Mr. H. Collard. In the interval, awaiting the planting season, Mr. Collard was sent to Fiji to gather first-hand information as to the history of the disease in those islands, its present status, and the possibility of securing resistant or immune stock.

Attempts to isolate any constant fungal or bacterial agent from the various parts of affected plants were unsuccessful, and after a period of six months, during which visits were constantly made to various parts of the affected area, noting the behaviour of plants in deserted plantations as well as the initial and early stages of infection in other plantations, it seemed highly probable, in view of discoveries mentioned later, that the trouble would not be attributed to any such agent.

Meanwhile the Investigation was also devoting attention to the matter of nematodes or eelworms, since these were found to be abundant in all plantations, and their presence readily indicated by the almost constant presence of galls on the roots of affected and apparently non-affected plants. Plants from northern Queensland were planted in tanks filled with steam-sterilized soil, and others in tanks of soil from an infected plantation, with a view to settling the possible effect of soil factors, e.g., nematodes, parasitic fungi, and bacteria, in a purely preliminary manner. It soon became apparent that such experiments, if they were to be of any scientific value, would have to be carried out under such conditions of control as obtain in a properly equipped glasshouse. This development and the necessity for a glasshouse for the purposes of other experiments, which were to be tried out, compelled the Supervisor to request the erection of such a glasshouse. The cost of the erection of this building was considered to be insignificant for scientific purposes where the national loss due to the disease was so great.

Keeping in view the possibility that the disease might be of the mosaic type of virus disease, attempts were made to produce the disease in healthy plants from northern Queensland, grown in sterilized soil,

and inoculated with the sap of affected plants. There was no evidence manifested for the transmission of the disease in the material used in these preliminary experiments, but it was recognized that further work along these lines should be carried out when the glasshouse was available. Similarly, no results were obtained by inoculating sterilized soil with three species of fungi, which were isolated from the roots of some specimens of plants affected with Bunchy Top. Further, the association, over a long period, of diseased and healthy plants in sterilized soil, and in the same pot, out-of-doors, so that the roots were in intimate association, failed to produce the disease in the healthy plants (an experiment which was later repeated with several plants under glasshouse conditions with the same results). Meanwhile, considerable work was accomplished on the experimental plots.

One of these with excellent soil had previously been affected with Bunchy Top, and, after being leased, had been thoroughly ploughed (part of it had rested for a period of several months) prior to the planting of the same by the Investigation in October, 1924. The plants were selected from various areas—some were plants which appeared to have stood out against the disease in affected plantations; some which appeared to have shown some signs of healthy constitution, taken from a deserted plantation; healthy plants from Bribie Island and Bracalba, Queensland; and three species of wild bananas from the neighbourhood of Cairns. The objects in this experiment were to determine the possibilities of resistance in local stocks, the possibilities of recovery from the disease, to make an intensive study of the incidence of the disease in originally undoubtedly healthy plants, and to determine the possible effects of cultural treatment in upholding resistance in healthy plants, and the possible resistance of wild stock to the disease.

Another plot of virgin land which had grown lantana for eight years was cleared, and planted with healthy stock from Bracalba. It was considered that, if soil factors such as physical and chemical constitution, bacteria, fungi, or nematodes played the major rôle in the direct production of the disease, it was possible that the plants on the plot of virgin land would stand out in contrast to those on the other plot which had carried Bunchy Top plants.

The early outbreak of disease in the plot of virgin land in January, 1925, and its rapid development throughout that plot within a few weeks, contrasted with the very slight development of the disease on the other plot, and led to an intensive study of the conditions prevailing in this plot. It was noticed that aphides were particularly abundant on this plot, an abundance possibly to be attributed to the topography of this lower plot being responsible for the bringing about of conditions more favorable for the aphides.

At once aphides were transferred from affected plants to healthy plants growing in sterilized soil, under insect cages, in the grounds of the laboratory, and the disease made its appearance in these experimental plants in about three weeks. The experiments were repeated by transferring aphides from affected to healthy Bracalba plants, grown under insect cages, in the University grounds, at Brisbane, where the disease appeared in two plants within a fortnight. Thus a definite lead was established to the investigation, and at once preparations made for following up these results in the glasshouse, arrangements for the construction of which had then been completed.

### 2. HISTORY OF THE DISEASE IN AUSTRALIA.

The earliest definite record from Australia of a disease in bananas, which can be regarded without doubt as that now known as Bunchy Top, is from a centre in the Tweed River area about thirteen years ago. There are growers who are of the opinion that the disease occurred much earlier in Australia, basing their opinion on statements about a "banana trouble" in the Clarence River area, or, as more often appears to be the case, on some malady affecting sugar cane in that region. There are no reliable data to support such opinions, and, in fact, the knowledge we now possess of the rate and mode of distribution of the disease detracts from the reliability of such statements. At all events, it can be definitely stated that the disease had been detected in the Tweed River area in 1913, although its status as a plant malady was not then recognized, the first interpretation being that we were concerned with a malformation due to non-parasitic agencies such as seasonal or climatic factors. It is of interest to note that the disease first made its appearance in a certain plantation from which stock was supplied to neighbouring planters, and this practice was continued until the extension of the disease within the original plantation indicated to the owner that he was viewing something more serious than a mere malformation of plants.

When the Advisory Committee, appointed in 1924 to inquire into the position in respect of Bunchy Top, visited the affected area in February of that year, it was learned from numerous growers who gave evidence before the Committee that "it was generally believed that the disease had emanated from the neighbourhood of this particular plantation." The reliability of this evidence is enhanced when an endeavour is made to discover the source of supply for the stocking of plantations—quite a large proportion can be traced to the neighbourhood of the plantation mentioned above. The fact that the trouble was brought under the notice of the Department of Agriculture, New South Wales, when the disease first appeared, enables the date of the earliest occurrence to be definitely stated. The fact that the history

of the disease has a very direct bearing on the results obtained by the present Investigation and on the recommendations made, renders such statements of some importance. The origin of the disease in the plantation referred to was, in all probability, Fiji, where the disease has been present since about 1885. This statement is based on the fact that banana stock was imported from Fiji and planted in the neighbourhood of the spot where the disease was first recorded in 1913. The identity of the Fijian disease is beyond dispute, since the visit of the Horticulturist connected with the present Investigation to Fiji in 1924.

There was a marked extension of banana-planting from 1913 onwards, and in New South Wales production reached its maximum in As planting progressed over the Tweed area, the disease gradually spread, this being facilitated by the distribution of affected suckers which, as we know, would serve to disseminate the virus in each locality. During the early years after the first appearance of the disease, Cabbage Top or Curly Top, as it was then known, was not considered such a dire malady as at present, but it gradually secured a thorough grip of the banana area which was still attracting a large number to the industry. The distribution of the disease by planters eventually led to the acknowledged dominance of the disease, particularly in certain spots within the Tweed area. During the past three years banana growing within the Tweed area has become a risky proposition, and since 1922 the industry has gradually declined. The disease became very well established at Terranora, and eventually wiped out the industry in that locality. There are to-day very few plantations around the Tweed where banana growing is maintained on a profitable basis. In a few places attempts have been made during the past three years to wrest some profit from banana land by planting on a sufficiently extensive basis to ensure some return, but such plantations have during the past two years become more and more intensely affected.

The position in the Tweed area at the present time is such that banana growing should be discouraged. Where the land was of a nature that it could be utilized for other purposes, the growers have gone over to some other means of livelihood, but in very many cases the plantations have been deserted, or are allowed to hold on, yielding a slight return, which, however, is becoming smaller each half-year.

In the Mullumbimby area the disease has become well entrenched, and there is an abundance of deserted plantations. In certain parts special efforts are made by removing all affected plants to fight the disease, with some measure of success. The proximity of deserted plantations renders such a task very difficult, but in some cases isolation renders considerable assistance. The origin of the disease in this zone can be traced to stock from the Tweed area.

The disease reached Bangalow in 1922, despite the institution of a regulation forbidding the passage of suckers from the north across a natural buffer zone in the neighbourhood of Byron Bay. The disease has been known along the Richmond River since 1923. There are many plantations along the Richmond where the disease has never appeared, others in which only a few plants have shown the disease during the past two years. This condition can be seen in such localities as Wardell. In the neighbourhood of Alstonville we have seen plantations in which comparatively few Bunchy Top plants have made their appearance, and by careful removal of these diseased plants the disease has not affected the industry in any way; in other cases there is a slight occurrence of Bunchy Top, but the owners, obsessed with the idea that the disease cannot be fought, have allowed the diseased plants to remain, thus eventually sealing the fate of the plantation. While we are not in possession of the details, which can be made available only as the result of a serious and thorough effort to obtain a census which will reflect the state of affairs in all the banana plantations, it is quite time to state that the area in the Richmond basin, which would embrace Wardell, Rousmill, and Alstonville, can be regarded as a lightly affected area, and one which, by the discharge of the recommendations made later in this report, could be conserved.

In proceeding from Alstonville to Tintinbar, via Teven, we saw plantations which have "gone out" with the disease. It is interesting to note that from Teven quite a number of plantations along the Richmond River received their stock, but we could not succeed in obtaining reliable evidence bearing on the ultimate source of affected stock at Teven. Occurrences of the disease in varied stages of intensity are to be noted between Tintinbar and Bangalow. Here and there on that stretch, as well as between Byron Bay and Mullumbimby, are to be met heavily affected and lightly affected plantations. No doubt good luck in respect of the original selection of suckers and isolation have together played a part, but in no plantation in this tract is the disease completely absent.

There can be little doubt that, were the details of the history of the plants available in the various plantations, the distribution of the disease could in every case be readily explained in terms of diseased suckers and the migration of aphides. We have on record the occurrence of the disease in Chatsworth Island, in the Clarence River.

Reference has been made to the occurrence of the disease in the Mullumbimby region, where it is widespread. Yet a syndicate is there cultivating about 100 acres of bananas, immediately above the deserted Soldiers' Settlements plantations. At Terragon, about 12 miles from Murwillumbah, and on the slopes of Mount Warning, the disease was but sparsely represented when visited during 1924. Isolation is, apparently, responsible for this condition.

At Upper Burringbar plantations are to be seen in various conditions. Some are in a hopeless state, some deserted; others are still producing. In the latter case attempts are made to cope with the disease by spraying with nicotine sulphate, or by digging out affected stools. On the whole, it may be stated that the disease is gradually increasing, which, under any circumstances, could be expected owing to the proximity of deserted plantations, and the fact that early detection of the disease and immediate digging out of the plants are not regarded as matters of extreme urgency by the growers.

The Soldier Settlement at Bilambil has been described. It will thus be seen that a very large part of the banana-producing area in northeastern New South Wales has passed out of the industry, and that practically no portion of the area is free from the disease. Unless something is done to arrest the spread of the disease in the more lightly affected areas, the industry in those parts must, within a few years, suffer the same fate as the Tweed area.

In Queensland the first record of the disease was made at Currumbin in 1916 in several plantations which were stocked with suckers from the Tweed area. Bunchy Top was there recognized as a disease identical with that responsible for the epidemic which devastated Fijian plantations from 1885 onwards. The growers concerned recognized that replanting was necessary, and for this purpose obtained suckers from Samsonvale, Queensland. It is of interest and significance that no sign of Bunchy Top was observed in these plants for three years.

In the intervening years, however, suckers from the affected areas made their way into new plantations along the Currumbin-Tallebudgera zone, and the disease became firmly established. Towards the end of 1922 it was recorded that at Currumbin and Tallebudgera 112 plantations, representing 1,250 acres, were affected in varying degree.

The Queensland Government, towards the end of 1921, attempted to stem the advance of the disease by prohibiting the removal of any vegetative parts of the banana plant from the affected South Coast fruit district to any other part of Queensland. Consideration was given to the creation of a buffer area between the affected districts in Queensland and the plantations to the north of the Brisbane River, but this idea was considered impracticable in view of the heavy compensation involved and the fact that a natural buffer area had not succeeded in checking the spread of the disease in New South Wales. (In the present state of our knowledge we are justified in stating that, provided no diseased suckers had already passed across such a buffer area, further spread of the disease northwards would have been prevented by this procedure.)

Despite the widening of the area over which the restrictions on removal were exercised, the disease was recorded from Brookfield, north of the Brisbane River, in December, 1923. The nearest area of infection previously known was about 40 miles distant. There are now but few localities south of the Brisbane River, such as Beenleigh and the neighbourhood of Coomera, from which the disease is absent.

On 30th December, 1925, the disease was discovered at Rush Creek in a plantation at Moorina, just south of the Caboolture River, 25 acres revealed the presence of the disease in quite a large number of plantations, the maximum infection in any plantation being about 20 per cent.

In January, 1926, it was discovered that the disease was rampant in a plantation at Moorina, just south of the Caboolture River, 25 acres showing 100 per cent. infection. Immediately a proclamation was issued by the Queensland Government prohibiting the shifting of suckers from any plantation south of the Caboolture River. The state of infection of the plantation at Moorina suggested that the disease had been present in that locality for several years. As large consignments of suckers had been sent northwards from the Dayboro district and from Moorina as recently as December, 1925, every effort was made to trace the consignments during the period 1923-1925. It was found that such suckers had been sent to various centres as far north as Innisfail, and the outbreak of Bunchy Top in such centres was anticipated.

It has been found that all plantations so far examined, which received suckers from Moorina since October, 1923, are affected, and that the presence of the disease at one plantation in the neighbourhood of Kilcoy (since completely eradicated), at Beerwah (one plantation), Yandina (one plantation), and Innisfail (two plantations), can be traced to suckers from the Moorina or Dayboro areas. The consignments from Moorina have been traced in completeness, and all receiving plantations inspected, but the very heavy consignments forwarded by agents from the Dayboro area necessitate much heavier inspection work. Undoubtedly many of these consignments may have consisted of healthy suckers, but, nevertheless, it is to be anticipated that further records of the disease in previously unaffected areas wil be made in the near future. In the meantime, the Queensland Government is prohibiting the shifting of suckers in any part of that State, and this will continue until a thorough examination enables the Queensland Department of Agriculture and Stock to determine areas which can be regarded as definitely free from the disease.

#### 3. ECONOMIC EFFECTS OF THE DISEASE.

The economic effects of Bunchy Top may be considered from three aspects, namely, (1) the actual destruction of banana plantatious and the loss of a most lucrative primary industry in heavily affected areas; (2) direct financial loss to individuals, districts, and States; (3) utilization of heavily capitalized land for the purposes of other primary industries.

1. Destruction of Plantations.—In New South Wales practically the whole of the banana-producing country is comprised within the Tweed, Brunswick, and Richmond River districts. Banana cultivation is attempted in a small way only in the Clarence River district, namely, at Chatsworth and Palmer Islands. In the Tweed and Brunswick districts banana growing as a profitable industry has practically disappeared. There is probably no plantation in any part of these districts which is free from Bunchy Top. There are a few profitable plantations, but these are mostly situated in outlying parts of the districts, and are more or less isolated.

A recent survey of the Tweed and Brunswick districts by Mr. H. W. Eastwood, of the Department of Agriculture, New South Wales, substantiates the general conclusions at which the Investigators had arrived as the result of frequent traversing of the affected areas. The following figures are taken from a report furnished by Mr. Eastwood to the Department of Agriculture, New South Wales:—

- (1) There are about ninety (90) plantations still profitable, with an approximate area of five hundred and fifty (550) acres, including the Maroomba plantation (100 acres).
- (2) There are, in addition, about fifty (50) growers who are still marketing fruit in varied quantities from the remnants of plantations.
- (3) There are at least eight hundred (800) deserted plantations, with an approximate area of five thousand (5,000) acres.
- (4) Fully 90 per cent. of the area producing bananas in 1922 has gone out of production.
- (5) Every plantation in the Tweed and Brunswick districts is infected with Bunchy Top, the percentage in the individual plantation varying from 5 to 90 per cent.

This represented the condition at the end of December, 1925, since which time there has probably been further development of the disease, as it becomes more prevalent during January and February.

The Richmond River district is generally infected, but, with the exception of such places as Teven, Tintenbar, and Tuckombil, only to a slight extent. The industry is increasing in importance. There are a few deserted plantations, but most of these have been deserted on account of age. There are many plantations quite free from Bunchy Top, and many again that are kept free by the digging out of affected stools as soon as the disease appears. In the Wardell area many plantations have never been affected.

It is not possible to produce detailed figures for Queensland, but the following facts will serve to indicate the losses due to Bunchy Top. In south-eastern Queensland Bunchy Top has produced damage comparable to that in the Tweed and Brunswick districts. In 1922 there were over 100 growers at Currumbin occupying over 1,000 acres of banana land; at the end of 1925 only four (4) plantations were left in cultivation, and they are practically done.

In Tallebudgera, once a great producing area, the position is much the same as at Currumbin, and production has practically ceased. Production has fallen by about 50 per cent. at Brookfield; every one of the 40 plantations producing bananas is affected with the disease. There are a few young plantations recently planted which, when last examined, were free from the disease.

In the Nerang district the disease has not yet brought about the same damage, but it is present. The Logan district now produces the bulk of the bananas in the area south of the Brisbane River.

2. Financial Loss to Individuals, Districts, and States.—The value of the banana industry to Australia is, perhaps, too little known to the people of the Commonwealth, and, consequently, the actual financial loss occasioned by Bunchy Top to the banana producer, to certain districts in New South Wales and Queensland, as well as to these States and the Commonwealth, is hardly appreciated by those who have no direct concern in the banana-producing areas. It is necessary that the loss so far occasioned should be realized in order that a serious effort be made to provide every practical means of conserving this important national industry and ensuring the utmost endeavour to save unaffected areas.

The following statement of losses in individual districts will serve to illustrate the extent and nature of the losses. In 1922 there were over 1,000 acres under banana cultivation in the Currumbin (Queensland) area, finding employment for approximately 200 men and a good living for several carriers; at the present time only five men are making a living from this industry in this area.

The following table\* will serve to indicate the losses to the individual growers, to the Currumbin district, and to Queensland during the period 1922-1926:—

| 1922 | 102,000 | cases | 4,400     | tons   | Railway   | freight† | £20,000 |
|------|---------|-------|-----------|--------|-----------|----------|---------|
| 1923 | 56,600  | ,,    | 2,460     | 77     | 22        | 22       | 11,000  |
| 1924 | 17,700  | 22    | 770       | 25.1   |           | 22       | 3,500   |
| 1925 | 2 500   | 27    | 110       | ,,     | ,,        | 22       | 500     |
|      |         | + Erc | om Currum | hin to | Melhourne |          |         |

When the Advisory Committee visited this area in February, 1924, the examination of the books of one grower showed that he had produced, on a 14-acre plantation, 3,725 cases of bananas in 1921, 3,500 cases in 1922, and 1,670 cases in 1923. The plantation is now deserted.

It would be difficult for any one who has not visited these devastated areas to visualize the completeness of the destruction wrought in such

<sup>\*</sup> Supplied by Mr. D. McLaurin, late secretary of the Currumbin Fruitgrowers' Association.

short time by a plant disease. The happenings at Tallebudgera are comparable to those at Currumbin, and the same results will be attained in the Brookfield area within a very short time, since production has already been reduced by 50 per cent. during the past two years, and the disease is now rampant. There is every danger that a similar fate awaits areas further to the north, such as Dayboro, Moorina, &c., unless there is determined action on the part of growers and the Government.

The following figures, made available by Mr. E. M. Smith, of the Tweed Fruit-growers' Association, show the estimated increased production during the period 1919-1922 (due to further planting-up), and the effects of Bunchy Top during the period 1923-1925, in the banana-producing areas of northern New South Wales:—

| 1919         |       |     | 180,000 | cases. |
|--------------|-------|-----|---------|--------|
| 1920         |       |     | 200,000 | 22     |
| 1921         | • •   |     | 350,000 | 22     |
| 1922         | • •   |     | 460,000 | 22     |
| 1923<br>1924 | • •   | • • | 250,000 | 22     |
| 1924         | • • • | • • | 200,000 | 22     |
| 1020         |       |     | 110,000 | 22     |

Fully 90 per cent. of the area in northern New South Wales which was producing bananas in 1922 has gone out of production, but new areas have come into bearing. The effects of Bunchy Top are therefore greater than the figures show.

The above will serve to indicate the losses in New South Wales to the growers and to the Government in respect of railway revenue, income tax, &c.

The price paid for good banana land in the Tweed district during the peak years of banana production was exceedingly high. Although the lucrative position of the industry may offer some justification, these prices would appear to have been excessive. At all events, the original cost of land adds further difficulties to the position now created by the Bunchy Top epidemic, since much of the land is unsuitable for purposes other than banana growing. The figures obtained by the Advisory Committee in 1924 have been substantiated by frequent inquiries when visiting various parts of the Tweed area—bearing plantations having realized £300 per acre, and virgin land covered with lantana having changed hands at £150 per acre.

Bunchy Top has thus been responsible for the ruination of many who entered the industry before the disease had gained control of the banana plantations in the Tweed and Brunswick districts in New South Wales and the Currumbin and Tallebudgera areas in Queensland. A case is on record where one man is now maintaining a dairy farm on 200 acres of land which at one time was divided into banana farms, furnishing a very good livelihood for fifteen men. The loss of the

industry has affected not only those who are actively interested in banana growing, but also a large number of labourers and those concerned in haulage, the production of case timber, &c. The cumulative effect of these losses as reflected in the various town centres in the affected areas is a subject of common comment by the townsmen.

3. Utilization of Land for other Purposes.—The losses which accrue as the result of the inability to replace banana growing in the devastated areas by an equally lucrative industry are exceptionally great. The greater portion of the land growing bananas in the Currumbin, Tweed, and Brunswick areas in 1922 has gone out of cultivation, and, in many cases, has been deserted. There is no alternative industry which, on this heavily capitalized land, would give a return equal to that obtained from bananas. Owing to the steepness and stony nature of the greater part of the land, attempts to wrest some profit from holdings which have not been deserted have necessarily been confined to minor crops on small areas. Unless measures are taken which will again permit of banana growing in the affected areas, these losses will be of a permanent nature.

Some few plantations have been replanted with sugar cane, but the area where this has taken place has been limited to those plantations with frontages to rivers or creeks to enable haulage of the cane to the mill by water. Many growers have engaged in bean cultivation, in some cases with good success, but the seasonable occurrence of Black Spot or Anthracnose (Colletotrichum lindimuthianium) renders bean growing an undertaking which is subject to considerable risk. Such cultivation is also limited to land which can be ploughed or chipped; quite a large proportion of the banana plantations are on boulder-strewn slopes and offer insuperable difficulties in respect of cultivation.

In very many instances abandoned plantations are now covered with lantana or grass. The usual practice, when plantations become unprofitable, has been to desert them or to turn in stock to eat down the stools. Eventually such land reverts to lantana or grass, and in the latter case the land is in some parts used for dairying or grazing purposes. As the average area of deserted plantations varies from 10 to 15 acres, it is necessary for several plantations to be grouped together to obtain sufficient land for a profitable dairy farm. The number of areas in which this can be done is naturally limited, and such a dairy farm starts off with an excessive capital outlay.

Some growers have attempted the cultivation of passion-fruit, peanuts, pineapples, tomatoes, &c., but for various reasons, except in isolated cases, these crops have not proved successful, and could not come into cultivation on a district scale.

The position is, therefore, that unless banana growing can again be safely undertaken in the devastated areas, this large tract of country will never return to its once flourishing condition.

### 4. GEOGRAPHICAL DISTRIBUTION.

Bunchy Top has a comparatively wide distribution among bananagrowing countries. In Australia the disease is well developed in northeastern New South Wales and south-eastern Queensland, and is present also in isolated centres of the banana areas of North Queensland. As infected suckers were sent from Queensland during 1925 to the North Gascoyne district of Western Australia, there is little doubt that the disease is also present in that State.

In Fiji about 40 years ago, Bunchy Top threatened the bauana industry with extinction. The disease is still present there, but is not regarded as a serious menace.

Bunchy Top has probably been present in Egypt since about 1900. Falmy(6) reports that the disease is found in every province, but is most prevalent in Lower Egypt.

In Ceylon Bunchy Top made its appearance in 1913, and has since spread over a considerable area of the island. Dr. Bryce, who investigated the outbreak in Ceylon, reports that the disease is also present in the Bonin Islands.

During 1924 reports were received of a disease which was causing damage in abaca or manila hemp in the Philippines. The symptoms of this disease closely resemble those of Bunchy Top, and since Bunchy Top has been transmitted to manila hemp, it is probable that the two diseases are identical in nature.

Mr. J. Campbell, Mycologist, Department of Agriculture, Fiji, has recently reported the occurrence of Bunchy Top in the Ellice Islands, where it was introduced some years ago in infected suckers imported from Fiji.

### 5. PREVIOUS LITERATURE AND INVESTIGATIONS.

The available literature dealing with the Bunchy Top disease is not extensive. This disease, although comparatively widespread in its distribution among banana countries, has apparently assumed greatest potentiality in Australia. In all countries in which it has occurred, efforts have been made on the part of workers to determine the nature of its cause and means of transmission, and to devise methods of combating it. In spite of considerable endeavour, nothing definite regarding these aspects of the disease had been proved up till the time of the present investigation.

As far as can be ascertained, Bunchy Top first made its appearance and attracted notice in Fiji. Writing in 1912, Knowles and Jepson (1) state "that the earliest record of the presence of this disease in Fiji was in 1891, although, according to certain old residents in the colony, it was known ten years prior to this date. About 1894 the banana industry was severely threatened, and many planters were compelled to abandon their plantations."

Knowles and Jepson considered the disease to be caused by a fungus "the exact systematic position of which has not yet been ascertained." This fungus was never found except in conjunction with nematodes. The disease was most prevalent in low-lying and poorly drained land, although it also made its appearance on the higher slopes. The local name of "strangles" was given to the disease in Fiji. The symptoms of the disease are briefly described, a special mention being given to the decomposed condition of the roots and corm. It is suggested that the disease is possibly identical with the banana disease described by Massee (1903) under the title of Banana Disease (Marasmius semiustus, Berk. Curt.)," occurring in Trinidad and other West Indian islands. Reference is made to the resemblance of the abnormal condition of the roots of plants affected with "strangles" to the root symptoms of a banana disease prevalent in Alexandria, attributed by Looss and Foaden (1902) to the agency of nematodes. An indirect reference is made to the occurrence of aphides on bananas during the 1891 outbreak of "strangles" in Fiji. Regarding the identity of this disease with Bunchy Top, it may be stated that photographs of the condition, received at Kew over 40 years ago from the Governor of Fiji, show exact symptoms of Bunchy Top.\* This is also proof of the fact that Bunchy Top occurred in Fiji prior to 1891.

Nowell(8) reports that an eelworm (Tylenchus similis), was described by N. A. Cobb as the cause of a serious outbreak of a disease of bananas in Fiji in 1890-91. There is little doubt that Bunchy Top is the disease referred to.

In 1921 Bryce(2) in a paper on "The Bunchy Top Plantain Disease," states that Bunchy Top first appeared in Ceylon in 1913, and has since spread over a considerable area of Ceylon. Fiji, Australia, Egypt, and the Bonin Islands are listed as countries in which Bunchy Top occurs. The cause of the disease was undetermined. Bryce considered it improbable that nematodes caused the disease. A species of Rhizoclonia was found to be prevalent on both the finer and larger roots, and it was thought that this was a possible cause of the disease. A preliminary experiment to determine whether the disease was caused by a filterable virus gave negative results. Bryce attributed the spread of the disease to the dissemination of diseased suckers for planting-up purposes. He states that it is not known whether any of the Ceylon plantains are immune to Bunchy Top, but that in Fiji the Gros Michel or Jamaica banana is immune. Observations made by the horticulturist of this Investigation during a visit to Fiji do not bear out this Manila hemp (Musa textilis) is reported as being susceptible to Bunchy Top. No mention is made of the occurrence of the banana aphis (Pentalonia nigronervosa) in association with Bunchy Top plants.

<sup>\*</sup> We are indebted to Dr. E. J. Butler, Director of the Imperial Bureau of Mycology, for this information. During 1923 Dr. Butler visited the Bunchy Top-infected area of New South Wales.

Darnell-Smith and Tryon(3), reporting in 1923 on their conjoint investigation of the banana Bunchy Top disease, state that their experiments and observations indicate that theories attributing the cause of the disease to soil depletion, loss of vigour in the banana stock, soil acidity or climatic factors, cannot receive any measure of support. The investigation, so far as it had gone, had failed to reveal the presence of a fungus organism capable of causing Bunchy Top. This work was still in progress; nematodes, in that they were not invariably found to occur in association with the disease, were not considered the exclusive cause of the malady. The banana aphis (Pentalonia nigronervosa) is reported as having received notoriety as the communicator, or even the prime cause, of the disease. It is stated that "although general observations do not favour this explanation, a remedy involving this insect theory is being tested in the New South Wales portion of the infected area."

In a report issued in 1924 on the "Bunchy Top Disease of Bananas," in Australia, Darnell-Smith (4) gives a description of its symptoms and occurrence, and the results of his work on the disease. He states that his observations suggest that Bunchy Top is the result of a fungus or bacterial infection of the roots or corm. He accounts for the sporadic occurrence of the disease by the fact that "the infecting organism may act as a saprophyte or a parasite." He particularly suspected Fusaria and bacteria attacking the roots as the cause of the disease, and reports that several organisms had been isolated. The possibility of insects transmitting or causing the disease was considered by him. A preliminary experiment to determine whether the banana aphis (Pentalonia nigronervosa) was a vector of the disease is reported as having given negative results.

In view of the fact that the investigation of the Bunchy Top disease was undertaken in 1924 by the present workers, Dr. Darnell-Smith's work in connexion with the disease was discontinued.

A special Advisory Committee, appointed in 1924 to inquire into the position in respect of Bunchy Top in Australia, in its report (5) gives a description of the disease and its complexities, its history in Australia, its economic effects, and recommendations for the future investigation of the problem.

Fahmy(6), in 1924, in a bulletin on "A Banana Disease caused by a species of Heterodera," described a condition in the banana which we consider, from the nature of the symptoms and photographic evidence, as identical with Bunchy Top. Communications with the author have confirmed this belief. The most reliable symptoms of Bunchy Top, i.e., dark, broken, green streaks along the midrib and secondary veins of the lamina, are present in the diseased plants in Egypt. Fahmy attributed the condition to the agency of nematodes (Heterodera radicicola), stating that "when over 30 per cent. of the roots were attacked, the disease

was externally apparent, the plant being dwarfed, and growth sluggish." The disease is present in every province of Egypt, but is most prevalent in Lower Egypt. It is stated that the disease has probably spread from one locality to another through the sale of infected suckers. All varieties of the banana grown in Egypt are susceptible to the disease, but symptoms are most noticeable in the Hindi variety (Musa cavendishii).

Fahmy (1924), states the disease was first observed at Alexandria over twenty years ago. It had its origin in a few plantations, but soon spread, causing considerable damage. According to Fahmy, Dr. Alex. Preyer, in 1901, in a contribution to the Khedival Agricultural Society, attributed the disease to a parasitic nematode or eelworm.

### 6. HOST PLANTS: VARIETAL SUSCEPTIBILITY.

Investigations carried on thus far have not shown any plant, other than a member of the genus Musa, as a positive host of the Bunchy Top disease.

All varieties of the banana grown in Australia are susceptible to Bunchy Top. The chief and only commercial variety of banana grown in north-eastern New South Wales and southern Queensland is the Cavendish (Musa cavendishii). This dwarf variety is very susceptible to Bunchy Top, but appears to be the only variety which can be grown profitably in these districts on account of climate and windy situations of plantations. Other varieties, grown in these areas to a minor extent, which succumb to Bunchy Top, are the Gros Michel (Musa sapientum), the Lady's Finger, and the Sugar variety. Two local varieties—the Hansonian (?) and the Vernon—also take the disease. In Fiji, the Cavendish, Gros Michel, Vi-ma-ma, and the Sawaqa (Musa fehi) are reported as susceptible to Bunchy Top. Three indigenous species of banana occur in the dense scrubs of north Queensland. These produce an inedible seed-bearing fruit. One of these species (Musa Banksii) has been successfully grown at Tweed Heads, and has been infected with Bunchy Top; the other two did not survive transplanting. Bryce (2) reports that, in 1918, in Ceylon, a plot of manila hemp (Musa textilis) was attacked by Bunchy Top. Bunchy Top has been transmitted to healthy manila hemp plants under glasshouse conditions at Tweed Heads.

A seed-bearing variety of banana from the Philippines (the F1 of a cross between  $Musa\ sp.$  var. alinsanag and an unknown male parent) bas also been infected with the disease.

There is a marked difference in the susceptibility of the Cavendish dwarf variety as compared with the tall growing varieties such as the Gros Michel and Lady Finger—the Cavendish being by far the most susceptible variety. Owing to its dwarf habit, the abnormal changes induced by the disease are more striking in this than in the taller varieties. Unless otherwise stated, the studies and experiments recorded below have been made with the Cavendish variety.

Observations do not suggest that Bunchy Top will attack any plant outside the genus Musa. Attempts to transmit the disease to the closely related genera Strelitzia and Ravenala, and to Canna sp., including the arrowroot (Canna edulis), the potato (Solanum luberosum), and maize (Zea mays) have failed to give results. Should another species of aphis, less specific in its host relationships than the banana aphis, be found capable of acting as a vector of Bunchy Top, an extension of the host range of the disease may be made possible.

On two or three occasions the attention of the Investigation has been drawn to a pathological condition shown by maize growing in Bunchy Top infected plantations. The maize plants were stunted, and their leaves were somewhat upright and contorted. Although a cause could not be definitely assigned to this condition in maize, we do not consider, from a study of its symptomatology, that it has any connexion with Bunchy Top. However, further inquiry into this matter is desirable.

Our attention was drawn during September, 1924, to the condition of a number of individuals of *Canna sp.* in a plot of these plants in a garden in Murwillumbah. Quite a percentage of the plants appeared to differ in their habit from the other members in the plot, and appeared to show symptoms very suggestive of Bunchy Top.

This impression was kept in mind, and, as stated above, was tested out in an attempt to transmit the disease to Canna sp., with unsuccessful results.

Careful study of the occurrence of the disease in various parts of the world has not demonstrated that any species or variety of banana is immune or highly resistant to Bunchy Top. Reference is made later in this paper to the position of the disease, as it now obtains in Fiji, with respect to the various varieties of bananas grown in those islands.

### 7. COMMON NAMES OF THE DISEASE.

The name "Strangles" was originally given to this disease of bananas in Fiji. Later, it became generally known in that colony as "the banana disease." In more recent years the names "Cabbage Top," "Curly Top," and "Bunchy Top," which were originally applied to the disease at various times during its progress in Australia, have also come into general usage in Fiji. In Ceylon, the disease has evidently always been referred to as the "Bunchy Top" plaintain disease, while in Egypt no special common name has been given to the trouble. Of the various names which have been applied to this malady, the name "Bunchy Top," for the reason of its more general application and its descriptive nature, seems to be the most desirable.

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### 8. SYMPTOMS OF THE DISEASE.

The external symptoms of Bunchy Top have already been described in some detail by Darnell-Smith (3), and Bryce (2). A study of the symptomatology of the disease has been made during the present investigation, under varied field conditions, and in the glasshouse. This has revealed some points not noted by previous workers. A preliminary study has also been made of the internal symptoms of the disease.

1. General Habit and Behaviour.—The symptoms of Bunchy Top are very characteristic, and the condition is easily identified. There is little possibility of confusing its symptoms with those of any other banana disease.

A plant may show symptoms of Bunchy Top at any stage of growth, from its first emergence from the ground, or as a plant fully grown and throwing a bunch. The name aptly describes the disease; the leaves of a badly infected plant are bunched together at the apex of the plant to form a rosette (Plate 8, Fig. 2). Owing to the failure of the leaf stalks to elongate, the leaves stand more erect than normal. Infected plants are markedly stunted, there being little growth in height once the plant has taken the disease.

The disease is systematic in nature. Suckers obtained from infected stools and replanted are almost invariably diseased. Suckers which have received infection from the mother plant, before being removed from the parent stool, show symptoms somewhat different from those obtaining in plants which have received the inoculum from an external source.

We propose using the terms primary and secondary respectively to distinguish these types of infection.

The difference in symptoms between the primary and secondary condition is really one of intensity. A plant receiving secondary infection in its earliest stages of growth may later exhibit symptoms just as marked as those of a primary-infected plant, while a sucker which has received infection from the parent plant, and shown the disease while growing in situ, rarely differs very much from the secondary-infected type. Plate 9, Fig. 1, depicts the habit of a primary-infected individual. Where primary infection has been acute plants may not grow more than 9 inches to 2 feet in height. Growth is very slow, and plants never mature or produce fruit. The plants do not die, at least within one or two years.

The habit of a secondary-infected plant will depend on the amount of normal growth which has been made before the plant shows symptoms of Bunchy Top. An early infected plant will later show symptoms indistinguishable from those of a primary-infected individual, due to the withering of the older healthy leaves. A plant may exhibit normal growth up till the last leaf before bunching, and then in the last leaf

show the first symptoms of the disease. Such a plant would mature a bunch of fruit—often of marketable quality. All intermediate stages are found between these extremes. Plates 6, 7, 8 show stages of the development of the disease in a secondary-infected plant.

When symptoms of the disease are exhibited after the bunch has been formed in the interior of the base of the pseudostem, there is evident an effort on the part of the plant to throw the bunch. Owing to the leaf congestion and constriction at the apex of the pseudostem, the bunch may with difficulty get out, only partly do so, or may fail altogether to emerge. Plants are thus often seen with partly-emerged bunches of small stunted fruit at their apices, standing erect, or jutting out at right angles. Where the constriction has prevented the bunch from emerging, on splitting open the pseudostem, the true stem supporting the bunch is seen to be contorted, or even bent back on itself, due to growth pressure.

Inspections of plantations in the infected areas of north-eastern New South Wales and south-eastern Queensland show the disease to be present to varying extent. A few plantations are perfectly free, while many others have 95 to 100 per cent. of the stools showing infection. At the present time the greater number of the heavily infected plantations are abandoned.

In affected plantations, healthy and diseased stools are often seen separated from one another by only a few feet. In other plantations every stool may bear one or several Bunchy Top suckers. In its early incidence in a plantation, which has made healthy growth for some time, the disease occurs sporadically. As a rule, definite centres of infection are not evident at first, but the later spread of the disease can often be accounted for by the proximity of diseased stools. Where planting has taken place in a badly infected area, or where infected suckers have been used in planting, every plant may exhibit symptoms of Bunchy Top within six months of planting. Bunchy Top plants do not wilt or die; a well-grown plantation which has become badly infected, when viewed from a distance, may look remarkably vigorous.

It is observed that, even in a stool which can be considered badly infected, suckers grow up which, for several months, may appear perfectly normal. Such suckers, however, usually take the disease before reaching maturity. In isolated cases, such suckers have been observed to grow to maturity, remaining healthy, and producing a good bunch. A stool of any age, or a sucker at any stage of growth, may become infected with the disease. Thus, in any infected plantation, the disease is seen in plants of all sizes. Suckers which have been replanted from a badly infected stool almost invariably become Bunchy Top, even if they are not showing symptoms of the disease at the time of removal from the parent.

The set-back received by a sucker in the separation of it from its parent, and the replanting of it as a separate individual, appears to hasten the manifestation of symptoms of Bunchy Top in that sucker. In this connexion it has frequently been claimed by growers that injury to healthy stools by runaway boulders has "caused" the injured stools to throw up diseased suckers. The same has been said of injury induced by desuckering in a Bunchy Top plantation.

In the case of a comparatively lightly infected plantation which was damaged by a severe wind-storm in 1924, it was claimed that the uprooting of large plants by the storm was the indirect cause of the later appearance of a large number of suckers showing infection. Exactly what factors are concerned here we are not certain. It seems possible that, in Bunchy Top, the infective agent in its relationship with the host may, in certain cases, arrive at a point of equilibrium where other factors are necessary to upset the balance, and cause the appearance of symptoms.

In a large stool which has shown the disease for the first time in only one or two suckers, it is often possible to select a sucker from the opposite side of the stool which, on planting out, will remain healthy. This is possibly due to such suckers at the time of removal being outside the diffusion range of the virus (and having escaped secondary infection from other members of the stool).

In the normal banana stool the individual plants are in organic connexion with one another. Young suckers are, for some time at least, physiologically dependent on the parent plants, but it seems doubtful, from observations on the spread of the disease in a stool, if this relation is maintained altogether among older plants in the stool. Also the removal of plants, which have fruited, from a stool may separate it into two or more organic units. This would account for cases of the apparent failure of the disease to spread as a primary infection to the whole of the stool. However, the practice of the selection of suckers for propagating from any part of a stool which has shown even minor infection is strongly condemned.

The period immediately prior to the throwing of the bunch is very frequently the time at which the first symptoms of disease become evident. This period cannot be correlated with the prevalence of aphides. We are led to believe that a plant may carry infection for some time before showing external symptoms—the physiological activities associated with the production and throwing of the bunch apparently causing the plant to react to the infection.

A Bunchy Top plant does not recover, but slowly goes on producing abnormal leaves over a long period. A few cases have been observed where infected suckers arising from a stool, after throwing two or three abnormal leaves, again produced a few leaves which were practically

normal in character, with almost total absence of green streaks (described below as the most reliable symptoms of the disease). Such plants, however, always reverted to the Bunchy Top habit. This condition was observed in the early summer when growth was most rapid. (Such behaviour has frequently been construed by growers as evidence of the "recovery" of infected plants).

2. Leaf Symptoms.—The first external symptoms of Bunchy Top appear in the leaves of the plant. The normal leaf emerges from the centre of the pseudostem with the leaf-blade wrapped tightly around the midrib in the form of a rod or "pipe." The leaf remains tightly rolled until it has almost fully emerged, and then commences to unfurl more or less evenly along its whole length. While unfurling the leaf stands erect, and when fully unrolled the elongation of the leaf stalk carries the blade clear of the pseudostem, and the leaf gradually assumes a position approaching the horizontal, making room for the next leaf which is pushing up through the pseudostem.

In the case of secondary infection, it is in a leaf which has unfurled in this manner that the first symptom of Bunchy Top is usually observed. The first definite symptom of the disease is the appearance of irregular, nodular, dark green streaks about .75 mm. wide along the secondary veins on the underside of the lower portion of the leaf-blade, along the leaf-stalk, or along the lower portion of the midrib.

In the first instance one, two, or several of these streaks may be present. Usually others appear later in the same region. In character they may vary from a series of small dark green dots to a continuous dark green line with a ragged edge, an inch or more in length. (Plate 3, Fig. 1.)

The lamina of the normal leaf is of an even rich green colour. From the midrib prominent vascular strands run out more or less perpendicularly at intervals as main veins. The area between the main veins is lined by secondary veins which are even in colour throughout the leaf. The normal midrib and leaf-stalk are of an even pale green colour, and are covered with a whitish waxy bloom.

Usually the dark-green streaks are first seen in the leaf-blade, but may later appear in the midrib and leaf-stalk of the same leaf. In some cases of secondary infection, an earlier indication of the later appearance of green streaks in the lamina is seen. This occurs in the "pipe" or tightly rolled heart leaf. It takes the form of the appearance of a number of irregular pale whitish streaks (Plate 5, Fig. 1) along the secondary veins of the tightly rolled lamina when the "pipe" is about one-half emerged from the pseudostem. When these pale whitish streaks appear as the first symptom, the "pipe" shows a slight transverse wrinkling along its length. On unfurling, a leaf which has shown these early streaks, bears numerous dark-green streaks along the secondary veins of the lamina. In other respects this leaf may not differ from the normal preceding leaves.

When, as usually is the case, the first symptoms take the form of a few characteristic green streaks in the lamina, midrib, or petiole, the "first-symptom" leaf, except for these streaks, appears normal in size, shape, and behaviour (Plate 7, Fig. 1). In the following leaf, however, while the "pipe" is still unfurled, pale whitish streaks are seen along the secondary veins of the leaf blade, These vary a great deal in number, depending on the degree of infection. The "pipe" or heart leaf now unfurls slightly abnormally, beginning to unroll from the top region, giving the partly unfurled leaf a funnel-like appearance. In this leaf, when unfurled, dark green streaks will be found to be present along many of the secondary veins of the lamina, and several dark green dots or lines are seen along the midrib and petiole. This leaf will be smaller than normal, slightly chlorotic, and the marginal portion of the lamina will be wavy and slightly rolled upwards.

The presence of the characteristic broken dark-green streaks (Plate 4, Fig. 1) along the secondary veins of the lamina, or along the midrib or petiole, is the most definite and reliable symptom of Bunchy Top. These streaks appear as the earliest external indication of the disease, and together with all other symptoms are not later retrospective in leaves which have been thrown earlier than the "first-symptom" leaf. The dark green streaks are not apparent when viewing the dorsal surface of the leaf in reflected light. The leaf should be inspected from the under-side so as to allow light to pass through it.

Successive leaves as they are thrown become more abnormal. There is a slight retardation in the rate of growth. The heart-leaf unfurls prematurely, and is slow in completing the process; often another leaf has begun to unfurl before the preceding one is fully unrolled. The leaves may become progressively smaller in size or successive leaves may be of similar dimensions. (In normal growth each leaf is larger than the preceding one.) There is a reduction both in width and length of the leaves, this being more noticeable in the case of the lamina than in the midrib. The petiole does not elongate normally, thus the leaves stand more erect than in the healthy plant. Leaves thrown soon after infection are distinctly chlorotic in appearance, but this character does not persist except on the margins of the laminae. After several abnormal leaves have appeared, extreme congestion is apparent at the apex of the pseudostem. The leaves are seen to have lost their normal symmetrical arrangement around the pseudostem, and to have assumed the "rosetted" condition. This characteristic arrangement of the leaves of a Bunchy Top plant does not result until several weeks after infection, and is thus not an early diagnostic symptom.

In colour the mature leaf of a secondary Bunchy Top plant is of a slightly more yellowish hue than that of a healthy plant. In the primary Bunchy Top plant the position is often reversed. Owing to the dark green streaking being present along the greater number of

the secondary veins, the general colour of the leaf is sometimes darker than normal. The lamina of such a leaf usually has a light yellowish green margin (Plate 9, Figs. 1, 2).

In texture there is a difference between the Bunchy Top and healthy leaf. Whereas the latter is elastic and pliable, the petiole, midrib, and lamina of the Bunchy Top leaf are harsh and brittle, and snap readily when bent or crushed in the hand. There is a distinct rigidity and apparent resistance to wilting shown by Bunchy Top leaves. They are not nearly so readily shaken by winds as healthy leaves.

The surface of the lamina of a Bunchy Top leaf becomes markedly corrugated as it matures, due to the growth of tissue in the region of the main veins producing ridging on the dorsal surface, and troughing on the ventral surface. This prominence of the main veins is not a diagnostic symptom of the disease. It is often seen in the leaves of healthy plants growing under adverse, or even rank, conditions.

The margin of the lamina of the young Bunchy Top leaf is wavy and slightly upward-rolled at intervals along its length. In the older Bunchy Top leaf the lamina is not supported normally by the midrib; both sides tend to hang down so as to be nearly parallel with each other, the margin, however, remaining up-turned or rolled.

In primary-infected plants, symptoms of the disease are apparent as soon as the first leaf appears above ground. The leaves are small, rosetted (Plates 9, Figs. 1, 2), and numerous broken dark green streaks are seen in the petioles, midribs, and laminae. The margins of the leaves are usually highly chlorotic, and are slightly rolled upwards.

3. Below-ground Symptoms.—Abnormal changes are seen in the root system of a Bunchy Top plant in an advanced stage of the disease. These symptoms we consider to be of a secondary nature; they are apparently due to the loss of immunity on the part of the roots of an affected plant, to the attacks of otherwise harmless soil organisms. This conclusion has been arrived at after considerable study of the root system of Bunchy Top and healthy plants. A good deal of attention has been given by previous workers to the presence of decaying roots associated with the Bunchy Top condition. The impression has been held that the disorganization of the root system by such agents as fungi, bacteria, or nematodes, was primarily connected with the causation of the disease.

The root system of the healthy banana consists of white, fleshy roots, cord-like in appearance, which spread through the soil in all directions. They are often of great length. From these cord roots arise large numbers of fine lateral roots which branch profusely. The lateral roots function only for a comparatively short time; new laterals are continually being produced as the cord roots spread through the soil.

In Bunchy Top plants some time after infection, many of the large fleshy roots are decayed and dead, while many others show purple patches upon their surface. In an advanced stage of the disease many of the roots have rotted back to within a few inches of the corm. All the lateral roots are either dead or purple in colour. On apparently healthy plants in the Tweed River district, many roots are found that are dead and decayed, and the laterals of many roots are purplish in colour. The purpling of the roots in the case of the banana appears to be a natural reaction to injury of any type, and to normally precede natural decay of laterals which have ceased to function.

The root system of a Bunchy Top plant, examined soon after the appearance of first symptoms of the disease, shows little difference in the number of decayed roots present, as compared with a healthy plant of the same age. As the disease progresses more and more, decay becomes apparent in the root system. However, Bunchy Top plants are sometimes seen with comparatively good root systems. Plants growing under glasshouse conditions in soil which has been sterilized, examined four months after the development of Bunchy Top, showed a relatively small difference in the number of decayed roots present when compared with healthy plants of the same age growing under similar conditions. (Plate 10 shows the root system of one of these plants.)

As the roots of a Bunchy Top plant rot back towards the corm, new roots, at first healthy in appearance, arise from the corm. On these an abnormal number of laterals is borne. After a time these laterals become purple in colour, and decay may be set up in these new roots. The Bunchy Top plant appears to be always endeavouring to maintain a scanty root system, but difficulty seems to be experienced on account of the attacks of decay-producing organisms in the soil. The disorganization of the phloem region of the vascular system, described later, may account for this loss of immunity on the part of the roots of Bunchy Top plants.

Galls caused by nematodes are seen on both the cord roots and lateral roots of Bunchy Top plants. The root system of practically every plant examined in the field has shown these galls to some extent. They may be numerous or only few in number. The surface roots are more often attacked than the deeper ones. Nematode galls appear to be just as plentiful in the field on the roots of healthy plants as on the roots of diseased individuals. Plate 10 shows a Bunchy Top plant grown in sterilized soil under glasshouse conditions, the roots of which are entirely free from nematode galls.

The surface of the cortex of a corm of a Bunchy Top plant in an advanced stage of the disease is often decayed. The corms of both healthy and Bunchy Top plants are in many plantations tunnelled by the beetle borer (Cosmopolites sordidus, Chev.). Where corms have been badly riddled by the beetle borer, decay in the region of the beetle

tunnels is common. The nematode-galling of the roots and beetle injury to the corm cannot be considered as contributing factors in the causation of Bunchy Top, and appear to be in no way connected with the disease.

4. Internal Signs of the Disease.—An internal macroscopic examination of the pseudostem and corm of a Bunchy Top plant reveals no pathological changes which, in our opinion, are of diagnostic value. In an advanced stage of the disease, extreme congestion of the leaf tissue is observed at the distal end of the pseudostem when split open. In suckers which have been topped for planting, it is not practicable, by inspection, to distinguish diseased from healthy individuals.

Both Darnell-Smith(3) and Bryce(2), in their descriptions of the symptoms of Bunchy Top, make mention of the vellowish or brown flecks or lines which run in all directions following the course of the vascular bundles in the corm and pseudostem, as symptoms of Bunchy Top. Observations made at Landsborough (Queensland), an area free from Bunchy Top, and on suckers from Bracalba (Queensland), from whence there has been no record of the disease, make it evident that the brown ducts cannot be considered as symptoms of Bunchy Top. Young healthy suckers are usually free from them. All healthy plants in a fairly advanced stage of growth, which have been examined, have shown these coloured ducts. Each vascular strand is accompanied on either side, for at least part of its length, by one of these canals, which vary in colour with the age of the sucker. They may be of the nature of continuous or broken lines, or have a monil form structure. They accompany the vascular bundles from the corm through the pseudostem into the midribs of the leaves. The coloured contents of these canals give a positive test for tannin with the Ferric chloride method, and are apparently solidified. The coloured ducts are particularly prevalent in a healthy corm where root decay or rotting of the corm surface has taken place. In a Bunchy Top corm which has been infected for any length of time, these coloured ducts are very noticeable, but young plants examined soon after infection may show few or none at all. The presence of coloured ducts associated with the vascular bundles cannot be considered as a symptom of Bunchy Top.

A microscopic examination of sections of tissue of the leaf system of an infected plant reveals a unique pathological condition of extreme interest. The following description is of a preliminary nature. It has not been possible to make a detailed study of the development of the condition, and future work is projected.

In the Bunchy Top plant disorganization has taken place in the phloem regions of the vascular system. Such alterations as are present are quite distinct in nature from the necrosis and lignification of phloem groups associated with the condition of leafroll of the potato (Solanum tuberosum), and are of value as a diagnostic internal

symptom of Bunchy Top. Studies made up to the present on this condition have been carried out on material fixed in Fleming's weaker solution and stained either in haematoxylin and crythrosin, safranin and light green, or in gentian violet and orange G. Healthy control material was obtained from a Bunchy Top-free area in Queensland and grown in the glasshouse at Tweed Heads. Comparative studies were made in hand sections and in microtomed paraffin sections  $12~\mu$  thick, of Bunchy Top and healthy material.

The external appearance of the broken dark green streaks in the lamina, midrib, and petiole of the Bunchy Top plant can be accounted for by the internal alteration in the region of the vascular bundles. The reliability of the appearance of dark green streaks as first symptoms of the disease is therefore evident.

In the secondary Bunchy Top plant internal alterations in the lamina, midrib, and petiole are only seen in the vascular bundles in the region of the dark green streaks, so that, sectious through these plant parts in the early stages of the disease show relatively few altered bundles. As the disease progresses, more and more vascular bundles show abnormal changes. In the primary Bunchy Top plant where infection is intense, every vascular bundle in the leaf is seen to have undergone alteration.

The vascular tissue of the banana consists of closed collateral bundles. From the corm, individual vascular strands run through the pseudostem into the midribs, where they branch profusely to form the parallel venation of the laminae; by branching and anastomoses the individual bundles communicate with one another. In transverse section, the normal vascular bundle is seen to be bordered on both phloem and xylem ends by a distinct sheath of fibrous elements. The xylem region is made up of spiral tracheides and xylem parenchyma. The phloem consists only of sieve tubes and companion cells.

A study of the internal pathological condition of the vascular system of a Bunchy Top plant is best made by cutting transverse and longitudinal sections of the midrib and leaf stalk of the primary diseased plant. Reference to Plate 19, Figure 2, will demonstrate how marked these alterations are. Plates 17-18 show individual healthy and Bunchy Top vascular bundles in transverse and longitudinal section.

A comparison of healthy and Bunchy Top bundles reveals the following alterations:—

(1) In the phloem region there is a suppression of development of the fibrous sheath. The region in which fibres are normally present is occupied by cells which are heavily laden with chromatophores.

- (2) The fundamental tissue in the neighbourhood of the phloem has become gorged with chromatophores. In the normal plant this region is almost entirely devoid of chlorophyll.
- (3) "Appositional" growth has taken place in the fundamental cells in contact with the phloem. The original cells have been divided up into angular cells by the growth of thin cellulose walls. Growth of these cell walls has apparently taken place in all planes. The newly formed cells contain numerous chromatophores and distinct nuclei.
- (4) "Appositional" growth of cell walls along all planes has taken place in the "sieve tubes" and companion cells of the phloem, giving rise to numerous angular cells along their length. These cells are uninucleate and their nuclei are of an abnormal type. The abundance of these nuclei in the phloem of a primary affected plant is a most noticeable feature in any transverse section and longitudinal section.

There is wide variation in the size and shape of these nuclei. Observations suggest that they are derived from normal nuclei by amitotic or direct division rather than by true mitosis. Although a binucleate condition of cells has not been definitely seen, numerous nuclei which appear to be undergoing division by constriction are observed. We recognize the necessity for further investigation of these remarkable changes in the phloem region of the vascular bundles of plants affected with this particular type of virus disease.

In general, the above description describes the alterations in the phloem region of the vascular bundle of a primary Bunchy Top plant as seen in the midrib. No alteration has taken place in the xylem region. The abnormal development of chlorophyll in the fundamental tissue is greatest in the region of the peripheral bundles of the ventral surface of the midrib, but is also present in association with the more internal bundles.

In the lamina of the primary Bunchy Top leaf, alterations similar to the above have taken place in both the main and secondary vascular strands. In some cases a reduction in the amount of sclerenchymatous tissue is also seen at the xylem end as well as the phloem end of the secondary vascular bundles. In the secondary Bunchy Top plant in the early stages of the disease, alterations of the vascular bundles are seen only in the region of the green streaks. Abnormal changes are not observed along the whole length of such bundles. This corresponds to the external appearance presented by an altered vascular bundle, i.e., as a broken dark green streak.

If a transverse section is made of a midrib which has shown dark green streaks as first symptoms of the disease, in the position of the streaks one or more vascular bundles in the ventral periphery of the C.18263.—5

midrib exhibit abnormal changes. "Appositional" growth has taken place in the fundamental tissue in the region of the phloem and for some distance from it. The newly formed cells are gorged with chromatophores. At this stage there is usually little alteration in the sclerenchymatous sheath, but the abnormal chlorophyllous tissue may extend as a band around the phloem end of the bundle. At this stage little or no alteration is apparent in the phloem itself. In a transverse section of the veins of the lamina of a leaf which is showing first symptoms of the disease, a similar condition is seen in the region of the green streaks. There is a proliferation of the mesophyll cells adjoining the phloem, with excessive development of chlorophyll. A strand of fibrous cells usually persists on the outer margin of the phloem end, so that a band of chlorophyll-bearing cells may extend on the inside of this fibrous strand, through the bundle from one side of the phloem to the other.

The brittleness of the leaf tissue of a Bunchy Top plant may, to some extent, be due to the suppression of development of fibrous tissue. Other factors, however, probably also contribute towards this characteristic.

The distribution in the plant of phloem alterations is subject to a good deal of variation. This is dependent on the stage of development of the disease. In the early stages of the disease a cross section of the midrib may show only a single altered vascular bundle. At a more advanced stage the greater number of the vascular strands are abnormal, while in the primary Bunchy Top plant every bundle has undergone change. In the primary Bunchy Top plant typical alterations are observed in the lamina, midrib, and petiole. In the pseudostem proliferation of the phloem and surrounding fundamental tissue, without the production of chloropyhll, has been observed as far down as the base of the pseudostem. The condition has not been observed in the corm or roots. Where a Bunchy Top plant has thrown a bunch, the true stem subtending the bunch, on examination exhibits typical alterations at the distal end. Around the fibres of the phloem region of many of the more peripheral vascular bundles, some of the cortical cells have undergone "appositional" growth to produce angular chlorophyll-bearing cells. Often a bundle is seen almost entirely surrounded by chlorophyll-tissue. Typical alterations are also seen in many vascular bundles in the rind of the fruit thrown by a Bunchy Top plant. These alterations do not produce an external appearance of green streaking in the fruit.

### 9. CAUSE AND TRANSMISSION OF THE DISEASE.

When the present Investigation commenced operations in May, 1924, one of the main problems before it was to determine the cause and means of transmission of Bunchy Top. Regarding this phase of the

disease, nothing definite had been proved by previous workers, either in Australia or other parts of the world where Bunchy Top occurs. Sufficient evidence was available from the work of Darnell-Smith and Tryon, as well as from the history of the disease in Fiji, to provisionally negative the suggestions which had been put forward that Bunchy Top was due to—(a) climatic factors; (b) "running out" of stock; or (c) chemical deficiency of the soil. Other theories relating to the cause of the disease which, in our opinion, had to be proved or disproved, were—

- (i) That Bunchy Top was due to parasitic bacteria or fungi in the soil.
- (ii) That nematodes were the causal agent of the disease.
- (iii) That Bunchy Top was a virus disease.
- 1. Parasitic Soil Factors (Bacteria, Fungi, Nematodes).—This factor was given first consideration for the reason that previous workers were inclined to give some credence to the idea that a soil parasite was the cause of the disease, and because the evidence of root decay associated with the Bunchy Top condition seemed to indicate the possible association of parasitic fungi and bacteria. Extensive observations were made on the root system of diseased and healthy plants in the Tweed River district of New South Wales, and on healthy plants in an area free from Bunchy Top at Landsborough (Queensland). Plantations in every stage of infection were examined, and evidence was collected with regard to the incidence of the disease. The early observations were made during the dormant period of growth, but from December, 1924, it was possible to study the first incidence of the disease in plants which had been brought to the Tweed River district from a disease-free area in Queensland.

In every plantation one was struck by the almost universal association of root decay with both healthy and Bunchy Top plants. This root decay was most marked in Bunchy Top plants in an advanced stage of the disease. The intense purpling of the surface of young roots of both healthy and recently diseased plants was suggestive of parasitic action, and received considerable attention.

Close observations were made of Bunchy Top plants for evidences of bacteria or fungi. As soon as facilities permitted, cultural work was commenced at the field laboratory at Tweed Heads with a view to isolating an organism as a casual agent of the disease. Extensive tissue platings were made of leaf, pseudostem, corm, and root tissue of diseased plants in every stage of disease on nutrient beef agar, potato dextrose agar, malt extract agar, prune juice agar, banana corm agar, banana leaf extract agar, gelatine and potato slopes. Studies were also made on inoculation of bouillon with infected tissue. It was possible to work on material within one or two hours after removal of

the plants under examination from the soil. Internal leaf and pseudostem tissues of diseased plants were invariably found to be sterile. Microscopical examination of diseased leaf tissue revealed a distinctly abnormal condition of the phloem region of the vascular bundles. There was, however, no evidence of the association of fungi or bacteria with this condition of the phloem, either under the microscope or when material was submitted to a fair range of isolation tests.

Later, attention was focussed on the decay of the root system and the surface of corms of Bunchy Top plants. Large numbers of platings on various media were made of decayed roots and corm tissue of plants in every stage of the disease, and also of healthy plant parts as controls. Isolation work extended over a period of six months, and, in spite of every endeavour, no positive results were secured. Two species of Fusarium were quite commonly isolated from decaying roots and the decaying areas of the cortex of corms of Bunchy Top plants. Platings of decaying roots from apparently normal plants also yielded both these species of Fusarium. Numerous species of bacteria were isolated from the decaying margin of corms and from decaying roots of Bunchy Top plants. No particular species of bacterium was associated with any degree of constancy. Their rôle seems to be that of secondary organisms. From the purple lateral roots of both healthy and recently diseased plants, both species of Fusarium were frequently isolated. When grown on potato dextrose agar, pure cultures of single spore strains of one of these species of Fusarium produced an intense blue colouration of the medium after about ten days' growth. The relation, if any, between this pigment production and the purpling of young roots was not determined. In older stools which had taken the disease, a species of Rhizoctonia was often found in association with decaying roots, both independently or in conjunction with Fusarium. In no case was the constancy of isolation sufficient to justify a suspicion of any of these types as a causal agent. At this stage, in view of results obtained from preliminary insect-transmission experiments, isolation work was

However, for observational purposes, soil infection experiments with two species of Fusarium and Rhizoctonia sp. were carried out. A pure culture of Rhizoctonia and pure cultures of single-spore strains of both species of Fusarium, were prepared as soon as possible after isolation from decaying roots. Prolific growth of each species of fungus was obtained on sterile puffed wheat, and heavy inoculations of each species were made in duplicate into autoclaved soil contained in fourteen-inch earthenware pots. Six days after inoculation, on the 12th February, 1925, young healthy suckers from a disease-free area in Queensland were planted in these pots, one to each pot. Before planting, each sucker was surface-sterilized in 1-in-1200 corrosive sublimate. Two pots of autoclaved soil were planted with similar suckers as controls.

All plants in the experiment, as well as the control plants, made only poor growth owing to cold weather. When this experiment was terminated, in October, 1925, both inoculated and control plants were free from Bunchy Top. An examination of the root system of all the plants was made, but this did not indicate that any of the fungi worked with were parasitic towards healthy plants.

2. Soil Sterilization.—Concurrently with isolation work, and as early as the growing season would permit, a pot experiment along the lines of soil sterilization was set up during 1924 in the open, at the laboratory at Tweed Heads, with a view to throwing some light on the possible association of parasitic soil factors with the disease. It was reasonable to believe that if the disease was caused by certain types of parasites, soil sterilization should control the trouble. The grounds of the laboratory at Tweed Heads are situated about 4 miles from the nearest banana plantation.

This experiment was conducted in eight No. 24-gauge corrugated galvanized iron tanks, 3 feet in diameter and 2 feet deep. These tanks were filled with soil from a badly infected plantation, the soil having been dug from the neighbourhood of rank Bunchy Top stools. The soil in four tanks was sterilized by means of steam. For this purpose use was made of the apparatus shown in Plate 11, Fig. 2. Steam at a pressure of 30 lb. per square inch was applied through the fourpronged pipe attachment, which was inserted in each pot of soil. The lower 10 inches of each prong of this attachment consisted of fine brass gauze tubing. With the apparatus in position, the soil in each pot was covered with sterilized corn sacks, and each pot was steamed for two and a half to three hours. After steaming for about one and a quarter hours a temperature of 100° C. was registered at all points on the surface of the soil, the lower portion of the soil being first raised to high temperature, the rising steam then heating the soil above. At least a temperature of 100° C, was maintained throughout the soil for one and a quarter hours.

On the 4th November, 1924, one week after sterilization, healthy suckers from a disease-free area in Queensland were planted in both sterilized and unsterilized soil—one sucker to each pot. Before planting, each sucker was surface sterilized by dipping completely in 1 in 1200 corrosive sublimate for two hours. The four pots of unsterilized soil were arranged as far away from the pots of sterilized soil as the grounds of the laboratory would permit, and each pot in each group was separated by a distance of 6 feet. This experiment was conducted in the open without insect-cages. It was thought that the isolation of the laboratory grounds would, to some extent, control the factor of insect transmission. All plants in both sterilized and unsterilized soil made vigorous growth from the commencement. From the 14th February, 1925, till September, 1925, at weekly intervals each plant received a thorough spraying with a 1 in 800 solution of Black Leaf 40.

In August, 1925, nine months after planting, all plants in both sterilized and unsterilized soil were healthy, and had made equal growth. On the 26th August, 1925, one of the suckers from a plant growing in a pot of sterilized soil showed first symptoms of Bunchy Top. On this date, all the remaining plants in the experiment, including those growing in the pots of unsterilized soil, were still healthy. This position was maintained when this experiment was terminated in October, 1925. The behaviour of the plant growing in sterilized soil is intelligible in the light of the results of other experiments recorded below. On several occasions during spraying operations, banana aphides (Pentalonia nigronervosa) were noticed in this, and later (in September, 1925) on two of the other plants growing in sterilized soil. These plants were in close proximity to caged aphis-infested plants belonging to another experiment.

In view of the fact that healthy plants, growing for eleven months in soil obtained from the proximity of Bunchy Top stools, remained free from the disease, we are able to conclude that the soil does not carry infection. (The soil used was probably free from aphides, as there was a lapse of ten days from the time of its removal from the plantation till suckers were planted in it.) An experiment described later, showing that the association of Bunchy Top and healthy roots in sterilized soil, in the absence of aphides, does not cause transfer of the disease, further emphasizes the fact that the soil in which Bunchy Top plants are grown does not serve as a medium for infection. An examination of the root system of the four plants growing in unsterilized plantation soil showed the presence of large numbers of nematode galls on the cord roots and laterals of all plants. Several roots on each plant were also showing decay. On the roots of the plant which developed Bunchy Top while growing in sterilized soil, no nematode galls were found, although several roots were showing decay similar to that of the plants growing in unsterilized soil.

This experiment helps to eliminate the possibility of nematodes or parasitic fungi or bacteria being concerned in the causation of Bunchy Top.

3. Ultra-microscopic Organism or Virus.—The possibility that Bunchy Top is caused by the association with the plant of an ultra-microscopic organism or virus received early consideration. The general nature of the leaf symptoms was suggestive of such a cause for the disease. Both Professor Stakman (Minnesota, United States of America), and Dr. E. J. Butler (England), who visited the infected area after the Pan-Pacific Congress in 1923, were agreed that Bunchy Top had many of the characters of a virus disease. Previous workers on the disease in Australia had, for the greater part, confined their attention to parasitic soil factors as the probable cause of the trouble. The possible virus-nature of the disease therefore called for further inquiry.

Dr. Bryce(2) records having conducted an experiment in Ceylon to determine if the disease was caused by a filterable virus, but reports having secured negative results.

During 1922, Mr. W. J. Marks, a banana-grower of Terranora, New South Wales, announced that he had discovered the cause and cure of Bunchy Top. He stated that Bunchy Top was due to aphides, which attacked the plants, and that aphides could be kept in check sufficiently to prevent Bunchy Top by spraying the plants at intervals with kerosene emulsion. Mr. Marks conducted an experiment to demonstrate the effectiveness of his treatment. He failed to prove that spraying would prevent Bunchy Top altogether, but it is now evident from the results after thirteen months' operation of the experiment, that he was securing partial control of the disease.

In 1923, Dr. Darnell-Smith (4) in a preliminary experiment conducted in Sydney, attempted to secure transmission of the disease by transferring banana aphides from Bunchy Top to healthy plants. This experiment gave negative results.

Evidence is presented below which serves to place Bunchy Top in the category of the virus diseases.

4. Juice Transfer Inoculations.—Two attempts have been made to transmit Bunchy Top from diseased to healthy plants by artificial inoculation. Juice was expressed from shavings of Bunchy Top corm, pseudostem and midrib tissue, and was separated from the pulp by straining through muslin. The undiluted juice was applied to healthy plants by hypodermic syringe injection, needle pricks, and mutilation of leaves along the veius. Endeavours made thus far to transmit the disease in this manner have failed to give positive results.

Experiment No. 1.—On the 4th November, 1924, eight banana suckers from a Bunchy Top-free area in Queensland were planted in sterilized soil in 3 feet x 2 feet corrugated iron pots—one sucker to each pot. On the 9th December, 1924, and again on the 12th January and the 24th February, 1925, each of four plants received inoculation of juice expressed from the leaves and corms of Bunchy Top plants in both early and advanced stages of the disease. Inoculations were made by hypodermic syringe injection, needle-pricks, and leaf mutilation into the younger leaf tissue and into the pseudostem and corm of each plant. The inoculations apparently induced no disturbance in the plants other than local necrosis of injured tissue. The growth of inoculated plants was equal at all stages to that of the control plants. From the 14th February, at weekly intervals, all plants in this experiment were thoroughly sprayed with Black Leaf 40 (1 in 800).

On the 4th June, 1925, one of the inoculated plants in this experiment showed first symptoms of Bunchy Top. Several individuals of *Pentalonia nigronervosa* had been observed on this plant during February, 1925, but were at once removed by spraying. During March,

aphides were again observed on this plant, and were removed. In August, one of the control plants showed symptoms of the disease. Aphides had been observed on this plant on three occasions. When this experiment was terminated in October, 1925, all the remaining plants were still healthy. This experiment was repeated later under insect-proof glasshouse conditions.

During April, 1925, an insect-proof glasshouse became available at the laboratory at Tweed Heads. The dimensions of this glasshouse (Plate 11, Fig. 1) were 42 feet x 22 feet. It was divided by an insect-proof glass partition into two sections—one 24 feet x 22 feet, and the other 18 feet x 22 feet. These sections are hereafter referred to as section A and section B respectively. All ventilators of the glasshouse were covered by fine brass gauze (50 meshes to the linear inch). All soil used in experiments in the glasshouse was sterilized by steaming. Before being used, both sections of the glasshouse were thoroughly funity gated to remove insects.

Experiment No. 2.—On the 27th August, 1925, four healthy Cavendish banana plants, which had been planted on the 16th April, 1925, in sterilized soil, in four large corrugated iron pots in section A of the glasshouse, each received inoculation of unfiltered expressed juice of a Bunchy Top plant in an advanced stage of the disease. The juice was obtained by pressure from shavings of corm, pseudostem, and midrib tissue. Juice-transfers were made to each plant by hypodermic syringe injection into the midrib and petiole, and by needle-pricks into and leaf mutilation of the younger leaves and the unfurled heart leaf. Hypodermic injections were also made into the pseudostem of each plant. From the corm of each plant a plug of tissue was removed with a flamed cork borer and about 2 c.cs. of infected juice were run in, the hole then being plugged with sterile cotton wool. Each plant again received similar inoculations on the 9th September, 23rd September, and the 8th October, 1925. At the time of the first application the plants were about 2 feet high, and had thrown either their tenth or eleventh leaf.

At the termination of this experiment on the 30th January, 1926, all inoculated plants and their controls were healthy and vigorous. Each plant had produced one or more suckers, which were also healthy. Apparently no effect on growth was produced by the inoculation. The average maximum temperature during the period following inoculation was 88° F. We do not consider that the phenomenon of masking would account for the absence of symptoms in the inoculated plants; it is during the warmer months of the year—December to March—that the appearance of the disease is most marked in infected plantations.

5. Preliminary Experiments on the Transmission of Bunchy Top by the Banana Aphis (*Pentalonia nigronervosa*).—Following on observations made early in January, 1925, on the incidence of Bunchy Top in

plants obtained from a disease-free area in Queensland, and planted at the experimental plots at Cobaki, New South Wales, a preliminary experiment was conducted to determine whether the dark banana aphis (*Pentalonia nigronervosa*) was capable of acting as a vector of Bunchy Top.

A healthy plant from Queensland, which had been growing for two months in a tank of sterilized soil in the open at the laboratory. Tweed Heads, was netted in with cheese-cloth. A large batch of banana aphides from a Bunchy Top plant in the early stages of the disease was transferred to this plant. The purpose of the cheese-cloth cage was to prevent aphides spreading on to neighbouring plants belonging to other experiments. The aphides multiplied rapidly. The date of transfer of the aphides to the healthy plant was the 17th January, 1925. On the 10th February, 1925, 24 days later, this plant was showing definite first symptoms of Bunchy Top. The control plant remained healthy till removed in October, 1925. Again, on the 15th February, 1925, aphides obtained from a Bunchy Top plant were transferred to another caged healthy Queensland plant, which had been growing in sterilized soil for three months. (More plants were not then available.) On the 13th March, 1925, 26 days later, this plant also showed symptoms of the disease, while the control plant remained healthy.

Two experiments conducted by Professor Goddard, Supervisor of this Investigation, furnish further evidence of the ability of the banana aphis to transmit Bunchy Top.

On the 15th February, 1925, aphides from Bunchy Top plants were placed on six young healthy plants from Bracalba, Queensland (a Bunchy Top-free area), growing in tanks of Tweed and Brisbane soil, two plants to each tank, under insect cages, in the grounds of the Queensland University. In one tank (Tweed soil) both plants showed first symptoms of Bunchy Top on the 28th February; in another tank (Brisbane soil), one plant showed symptoms on the 4th March, and the other on the 5th March, while in the remaining tank (Tweed soil), both plants were showing symptoms of the disease on the 16th March, 1925.

Again, on the 16th March, 1925, aphides from Bunchy Top plants were transferred to six young healthy north Queensland plants growing under similar conditions to those in the last experiment. All plants in this experiment were showing disease symptoms on the 12th April, 1925. The control plants to these experiments, including two growing in Tweed soil, all remained healthy till removed in November, 1925. The caged tanks in which these experiments were conducted were indiscriminately arranged, being separated only by 2 feet.

6. Final Transmission Experiment with the Banana Aphis (Pentalonia nigronervosa).—Healthy Cavendish banana suckers obtained from the C.13263.—5

Queensland Government Nursery, Bribie Island—an area where Bunchy Top has never occurred, were used in the final transmission experiment with aphides in the insect-proof glasshouse at Tweed Heads.

On the 16th April, 1925, 40 healthy suckers were planted in steam-sterilized soil in twenty 3 feet x 2 feet galvanized iron tanks in the glass-house. Twenty suckers were planted in ten tanks—two suckers to each plant—in section A of the glasshouse. The other twenty suckers were planted similarly in section B. On the same date, six healthy suckers were planted under muslin cages in two large tanks of sterilized soil—three suckers to each tank—in section B of the glasshouse. All suckers were of even size, and were free from the beetle borer (Cosmopolites sordidus). Each sucker, before planting, was dipped for one hour in 1-in-1,200 corrosive sublimate.

The soil used was a red volcanic loam, and although not of even quality in all the pots, was so arranged as to give adequate control of this factor. Each pot of soil was sterilized *in situ* in the glasshouse by steam, as described previously.

Owing to cool weather during the winter, the early growth of all plants was slow. On account of aspect the plants in section A of the glasshouse made more rapid growth than those in section B. By August, 1925, all plants were making healthy growth. The twenty plants in section A were slightly more forward than those in section B.

On the 29th August, 1925, approximately 150-200 individuals of Pentalonia nigronervosa obtained from two Bunchy Top plants, were transferred to each of the twenty plants growing in section B of the glasshouse. The method of transfer of the aphides is depicted in Plate 6, Fig. 1. A portion of leaf tissue of a Bunchy Top plant with a colony of aphides attached was placed on a piece of thick blotting-paper, resting on one of the leaves of each plant. Actual contact between Bunchy Top and healthy tissue was thus prevented. Each portion of tissue was carefully examined before transfer to the healthy plants, to make certain that no aphides other than Pentalonia nigronervosa, or any other insects, were being transferred. After the lapse of three days, the Bunchy Top tissue and blotting-paper were removed from each plant. By this time practically all the aphides had migrated to the healthy plants, and taken up a position on the sheathing bases of the leaves, or on the base of the pseudostem. The aphides multiplied rapidly. After a fortnight each plant was carrying a large colony of aphides, as shown in Plate 14, Fig. 1. These were clustered around the unfurled heartleaf, the sheathing bases of the leaves, and the base of the pseudostem. Very few aphides were seen on the surface of the leaf-blades.

The twenty control plants in section A remained free from aphides throughout the experiment.

An examination of the twenty aphis-infested plants on the 23rd September, 1925, revealed three plants showing definite first symptoms of Bunchy Top. On the 25th September, eighteen plants were showing symptoms of Bunchy Top. By the 29th September, 1925, all the infested plants were exhibiting definite symptoms of the disease. An examination on the same date of the twenty control plants in section A showed every plant to be healthy, and free from aphides. (After the appearance of symptoms of Bunchy Top, aphides were removed, or kept in check, by spraying the infected plants at intervals with Black Leaf 40.)

At the termination of the experiment on the 30th January, 1926, all the control plants were still healthy, while the infected plants were in a fairly advanced stage of Bunchy Top (Plates 12 and 13). Plates 6, 7, and 8 depict one of the infected plants at different stages in the development of the disease. Reference to the accompanying table will show details of the incidence of first symptoms in the aphis-infested plants.

The period from the time of placing aphides on the plants to the appearance of first symptoms varied from 23 to 29 days, with an average period of nearly 25 days. There was thus a fairly definite incubation period in the development of Bunchy Top in the plants in this experiment. Field observations, however, suggest that the incubation period may be influenced by a number of factors at present unknown. The age of the plant seems to exert some influence—younger plants apparently reacting to the disease more quickly than older individuals. We are therefore not inclined to believe that the incubation period of Bunchy Top, developing in plants of different ages and under varying conditions, is at all constant. Low temperature masking may have some effect in lengthening the incubation period during cool weather.

On 9th September, 1925, a large number of individuals of Pentalonia nigronervosa obtained from healthy plants growing at Landsborough, Queensland (a Bunchy Top-free area), was transferred to the six healthy plants (mentioned above), growing under insect-cages in section B of the glasshouse. The banana aphis is widespread through parts of North Queensland from which there has been no record of the disease. The object of this portion of the experiment was to demonstrate that non-virulent aphides, i.e., aphides which had not fed on Bunchy Top plants, were incapable of inducing the disease under similar conditions where positive results were secured with the use of local aphides fed on Bunchy Top plants,

Table showing details of incidence of first symptoms of Bunchy Top in plants infested with Pentalonia nigronervosa from Bunchy Top plants:—

| No.<br>of<br>Plant. | No. of Leaves<br>thrown at<br>time of<br>Infestation, | Date of Infestation. | Date of<br>appearance<br>of first<br>Symptoms. | Incu-<br>bation<br>Period. | No. of Leaves<br>thrown at<br>time of<br>appearance<br>of first<br>Symptoms. | Nature and Position of<br>first Symptoms.  |
|---------------------|---|----------------------|--|----------------------------|--|--|
| Iı                  | Five, sixth emerging                                  | 29.8.25              | 24.9.25  | 24 days                    | Eighth   | Several broken green streaks<br>on right and left side of<br>lower portion of lamina of  |
| I <sub>2</sub>      | Five, sixth   | 29.8.25              | 25.9,25  | 25 days                    | Eighth   | eighth leaf Pale whitish streaks apparent in unfurled "pipe" of  |
| $I_3$               | Six, seventh just showing                             | 29.8.25              | 25.9.25  | 25 days                    | Eighth   | eighth leaf Broken green streaks in petiole and on both sides of lower portion of lamina of eighth   |
| Ι4 .                | Six, seventh emerging                                 | 29.8.25              | 25.9.25  | 25 days                    | Eighth   | leaf Eighth leaf unfurling showing several green streaks in lamina and one green streak  |
| I <sub>5</sub> i    | Seven, eighth<br>emerging                             | 29.8.25              | 25.9.25  | 25 days                    | Ninth  | in midrib  Ninth leaf unfurling showing green streaks in midrib and  |
| Is .                | Seven, eighth<br>showing                              | 29.8.25              | 25.9.25  | 25 days                    | Ninth  | several in sides of lamina Faint-green streak in petiole, several faint-green streaks on left side of lamina of ninth leaf                               |
| I,                  | Six, seventh<br>emerging                              | 29.8.25              | 25.9.25  | 25 days                    | Ninth  | Two faint broken green<br>streaks on right side of<br>lower portion of lamina of   |
| $\mathbf{I}_{s}$    | Eighth, ninth<br>emerging                             | 29.8.25              | 25.9.25  | 25 days                    | Eleventh   | ninth leaf  Long broken green streaks in  midrib of eleventh leaf, none  |
| $\mathbf{L}_{0}$    | Five, sixth<br>emerging                               | 29.8.25              | 25.9.25  | 25 days                    | Eighth   | in lamina Eighth leaf unfurling showing two green streaks in midrib and several on both sides of lamina  |
| I <sub>10</sub>     | Six, seventh showing .                                | 29.8.25              | 23.9.25  | 23 days                    | Eighth   | Several broken green streaks<br>on right and left side of  |
| I <sub>11</sub>     | Six, seventh<br>emerging                              | 29.8.25              | 25.9.25  | 25 days                    | Eighth   | lamina, none in midrib Three faint-green streaks at base of left side of lamina,   |
| 1,2                 | Seven, eighth   | 29.8.25              | 28.9.25  | 28 days                    | Tenth  | none in midrib or petiole<br>Pale-whitish streaks in un-<br>furled "pipe" of tenth leaf  |
| I <sub>13</sub>     | emerging<br>Seven, eighth<br>emerging                 | 29.8.25              | 24.9.25  | 24 days                    | Tenth  | and right side of lamina of<br>tenth leaf, none in midrib or   |
| I14                 | Seven, eighth<br>emerging                             | 29.8.25              | 25.9.25  | 25 days                    | Ninth  | petiole Ninth leaf unfurling showing pale-geen streaks on both sides of lamina, none in midrib or petiole  |
| Ι <sub>15</sub>     | Eight, ninth<br>almost un-<br>fyrled                  | 29.8.25              | 24.9.25  | 24 days                    | Tenth  | Several broken green streaks in<br>petiole, none in lamina or<br>midrib  |
| I <sub>18</sub>     | Eigl.t, ninth<br>emerging                             | 29.8.25              | 23.9.25  | 23 days                    | Twelfth  | Pale-whitish streaks in un-<br>furled "pipe" of twelfth<br>leaf  |
| I <sub>17</sub>     | Eight ninth,<br>emerging                              | 29.8.25              | 29.9.25  | 29 days                    | Twelfth  | Several pale-whitish streaks in<br>unfurled "pipe" of twelfth  |
| I <sub>18</sub>     | Nine, tenth<br>emerging                               | 29.8.25              | 24.9.25  | 24 days                    | Twelfth  | Several pale-whitish streaks in<br>unfurled "pipe" of twelfth<br>leaf  |
| $I_{10}$            | Six, seventh<br>emerging                              | 29.8.25              | 24.9.25  | 24 days                    | Tenth  | Several broken green streaks in<br>both side of lamina, no   |
| I <sub>20</sub>     | Seven, eighth<br>showing                              | 29.8.25              | 23.9.25  | 23 days                    | Ninth  | streaks in midrib or petiole<br>Six broken green streaks in<br>lower portion of right side<br>of lamins of ninth leaf no<br>streaks in midrib or petiole |

Large colonies of aphides developed on the six plants on which aphides from healthy plants had been placed. At the end of two mouths all the plants were still free from Bunchy Top, but the

heavy infestation of aphides was beginning to cause injury to the plants. Large quantities of "honey-dew" had collected around the sheathing bases of the leaves and had fermented, causing a decay of the petioles. Three of the plants were therefore lightly sprayed with Black Leaf 40 to decrease the number of aphides. During December, 1925, the three unsprayed plants withered and died as the result of the attacks of the aphides. Up till the time of their death no indication of Bunchy Top was seen in these plants. Smaller colonies of aphides were maintained in the remaining three plants, and up till 30th January, 1926—that is, after four and a half months' infestation—they had not developed Bunchy Top.

This experiment definitely proves that banana aphides which have fed on Bunchy Top plants are capable of transmitting the disease to healthy plants. During the course of the experiment, no other species of aphis, and, with the exception of a few metallic green flies (Psilopus pachigyna Macq.), no other insect was found to be present in the glasshouse. Acarids (Tetranychus sp.) were noticed on the foliage of both infected and control plants during the latter part of the experiment.

7. Systemic Nature of the Disease—Sucker Transmission.—Field evidence collected during 1924 seemed to indicate that the incidence of the disease in many plantations had been due to the introduction of suckers from the areas infected with Bunchy Top. During 1924 and 1925 experiments were conducted to determine whether the disease was of a systemic nature.

Experiment No. 1.—On 7th November, 1924, ten small eyes were selected from a Bunchy Top stool in an advanced stage of the disease. Each eye was cleaned free from soil by scrubbing, and was submerged in 1-in-1,200 solution of corrosive sublimate for two hours. A plug of tissue was removed from each eye by means of a sterile cork borer and platings were made on potato dextrose agar and nutrient beef agar. The selected eyes were then planted in pots of autoclaved soil—one eye to each pot. In every case symptoms of the disease were evident as soon as the first leaf had spread out. After three months' growth each plant was showing the typical primary-infection symptoms. All plants during growth remained free from aphides. The tissue platings revealed no organism of interest.

Experiment No. 2.—On 20th April, 1925, twelve unemerged eyes were selected from plants in the field which had exhibited symptoms of Bunchy Top for only two to three weeks. It was considered that eyes which had not yet broken through would have escaped inoculation by aphides. The twelve eyes were surface-sterilized in 1-in-1,200 corrosive sublimate, and were planted in a 3 feet x 2 feet tank of sterilized soil in the glasshouse under insect-proof conditions. Eleven of the twelve selected eyes made growth. Each plant showed primary-infection symptoms from the time of its appearance above ground.

It is clear from these experiments that the disease can be spread by the propagation of suckers from infected stools.

8. Root Contact of Diseased and Healthy Individuals.—Pot experiments conducted to determine whether the contact of roots of healthy and Bunchy Top plants would cause transfer of the disease have indicated that in itself root contact is not a factor in the natural transmission of Bunchy Top.

On 6th November, 1924, one healthy sucker from a disease-free area in Queensland was planted in the same large pot of sterilized soil with three suckers from an advanced Bunchy Top stool. The experiment was conducted in the open at the field laboratory at Tweed Heads. The diseased suckers gave rise to primary-infected plants. Both diseased and healthy suckers were sprayed weekly with 1-in-800 Black Leaf 40. After eight months' growth under these conditions the healthy plant was still normal.

On 16th April, 1925, one healthy and three primary-infected suckers were planted in each of two large pots of sterilized soil under insect-proof glasshouse conditions. Each healthy sucker was thus surrounded by three infected ones. In January, 1925, the healthy suckers were still normal. One of the pots of this experiment is depicted in Plate 15, Fig. 1.

On the same date six healthy and six primary-infected suckers were planted in six 14-in. earthenware pots of sterilized soil under insect-proof glasshouse conditions. In each pot one diseased and one healthy sucker were pressed in actual contact under the soil. In January, 1926, all the healthy plants were normal. All suckers in these experiments were surface-sterilized in 1-in-1,200 corrosive sublimate previous to planting.

9. Transmission by Grafting.—An attempt to graft eyes from healthy plants on to diseased corms was made on 15th October, 1925. Six young eyes were removed from healthy plants and were each inserted on a diseased sucker in the position from which a similar sized eye had just been removed. Contact between the stock and scion was established with the aid of cord and adhesive tape. All other eyes were removed from the diseased suckers. The grafts were planted in a tank of sterilized soil under insect-proof glasshouse conditions. All the diseased stocks grew, but none of the inserted eyes appeared above ground. A later examination showed that they had rotted without making any growth.

The results of the above experiments definitely prove that Bunchy Top can be transmitted by the banana aphis (*Pentalonia nigronervosa*). There is definite evidence that the disease is systemic in nature. Microscopic examination and cultural tests have failed to indicate a bacterial,

protozoan, or fungal organism as a causal agent. As Pentalonia nigronervosa is widespread in banana plantations of North Queensland, where as yet no Bunchy Top has been reported, there is reason to believe that in north-eastern New South Wales and south-eastern Queensland this insect is carrying some factor which induces the disease. By analogy with other aphis-transmitted diseases, such as potato and tobacco mosaic, sugar cane mosaic, &c., it would seen that Bunchy Top comes in the category of the virus diseases.

This class we here consider to be constituted by those transmissible or infectious diseases, caused by an ultra-microscopic agent, which are perpetuated indefinitely by vegetative growth and propagation. Such diseases are in general characterized by chlorosis and may manifest other symptoms such as foliage distortion, excessive chlorophyll production, blight, stunting, proliferation, and internal lesions. Diseases caused by infective viruses fall into two divisions—one capable of being produced in healthy plants by artificial juice-transfer inoculation, as in the typical mosaic diseases, and the other in which juice inoculation is ineffective in producing the disease, as in the case of Leafroll of the potato. It is in this latter group of virus diseases that Bunchy Top should find a place.

There are many features of resemblance between Leafroll of the potato and Bunchy Top of the banana. Dwarfing, chlorosis, rolling, rigidity, and uprightness, the symptoms of Leafroll, are seen also in Bunchy Top. The phloem necrosis of Leafroll has a parallel in the phloem disorganization associated with the condition of Bunchy Top. Leafroll also resembles Bunchy Top in its inability to be transmitted from diseased to healthy individuals by artificial juice-transfer inoculation. This comparison is drawn only to further the evidence obtained as to the nature of the cause of Bunchy Top, and not as an inference that Leafroll and Bunchy Top are cognate diseases.

In view of the above, we feel justified in concluding that Bunchy Top is a transmissible disease caused by an ultra-microscopic agent. We are of the opinion that its chief vector is the banana aphis (Pentalonia nigronervosa), and that the spread of the disease in north-eastern New South Wales and south-eastern Queensland over wide areas, has been due first to the propagation of infected suckers, and then to natural transmission by aphides. There is evidence available that the soil, apart from its harbouring of aphides for a limited period, does not become infected. As attempts to transmit the disease by direct sap-inoculation have so far met with failure, it would appear that the disease cannot be spread by infected tools during cultural and harvesting operations, such as the pruning of suckers and the cutting of bunches.

It is possible that another species of aphis may be concerned in the natural transmission of Bunchy Top. On the lower leaves of young

banana plants a green species of aphis (probably Macrosiphum solanifolii) is sometimes found during the early summer These aphides do not remain on plants for any prolonged period. Two unsuccessful attempts have been made to arrange an experiment to test out the transmission capabilities of this species of aphis. These efforts have failed because the green aphides were unavailable when required. It is desirable that this species of aphis should be experimented with further.

10. Transmission of Bunchy Top to Abaca or Manila Hemp (Musa textilis).—Dr. Bryce(2) observed the occurrence of Bunchy Top in a plot of manila hemp in Ceylon in 1918. The exact identity of this disease with Bunchy Top in the banana was not established.

During 1924 reports were received from the Philippine Islands concerning a disease in abaca, the symptoms of which corresponded well with those of Bunchy Top in the banana. In October, 1925, definite evidence was secured by this Investigation of the transmission of banana Bunchy Top to manila hemp.

Through the courtesy of Dr. Darnell-Smith, six healthy manila hemp suckers were obtained from the Botanic Gardens, Sydney. Three suckers were planted in separate pots of sterilized soil in section A and in section B of the glasshouse. On 29th August, 1925, the three suckers in section B were infested with a large number of individuals of Pentalonia nigronervosa obtained from a Bunchy Top banana plant. The suckers had then thrown either their fourth or fifth leaf. On 6th October, 1925, all three suckers were showing definite symptoms of Bunchy Top. The incubation periods were 33, 34, and 38 days. The three control plants in section A remained free from aphides, and were still healthy at the termination of the experiment in January, 1926. Plate 14, Fig. 2, depicts one of the infected and one of the control plants.

The first symptoms appeared in the eighth or ninth leaf as broken dark green streaks along the midrib. The "first-symptom" leaf was otherwise normal in appearance and behaviour. The following leaf was slightly reduced in size and showed a distinct diffuse chlorosis; dark green streaks were present in the petiole and midrib but not in the lamina. All succeeding leaves were much reduced in size, and showed uprightness, rigidity, and slight chlorosis. The margins of their laminae were crinkled and upward-rolled; dark green streaks were present in the midribs and petioles, and faint green streaking was apparent along some of the secondary veins of the laminae.

As growth continued, the leaves assumed the rosetted condition, resembling more that developed by the tall-growing varieties such as the Sugar banana, than the dwarf Cavendish variety. The internal alterations in the region of the vascular bundles in the positions of the green streaks were apparently similar (in hand-sections) to those seen in the banana.

# 10. OBSERVATIONS ON THE BANANA APHIS (Pentalonia nigronervosa Coquerel).

The banana aphis (Pentalonia nigronervosa Coquerel) was first described by Coquerel in 1859, and later by Wilson in 1909. Froggatt(9), in 1923, contributed a paper to the Agricultural Gazette of New South Wales giving a description of the banana aphis, and mentioning that certain growers in the Tweed River district of New South Wales claimed that the presence of this insect indicated that the infested banana plants would later on be attacked by Bunchy Top. Coquerel describes (in translation) the banana aphis as follows:—"Of reddish brown colour, clearer on the head. Antennae, nervures, and cornicles blackish; legs yellow, with the base of the femora, tibia, and tarsi black." (After Froggatt.)

Baker (10), in 1920, gave the following description of the characters of the genus *Pentalonia:*—"Head with prominent antennal tubercles, which are, more especially in the apterous form, projected inward, gibbous and somewhat Myzus-like in appearance. Antennae of six segments, armed with sub-circular sensoria, the first segment gibbous like the antennal tubercles. Cornicles somewhat constricted near their middle, then again somewhat swollen near their distal extremity. Cauda rather small but elongate, subconical, slightly constricted about the middle. Fore wings with the radial sector extending abruptly downward and meeting the upper branch of the media with which it fuses but is diverted again towards its natural course near the tip of the wing. A closed cell is thus formed by the radial sector and the media, but at the margin of the wing there are the same veins as in the Aphidini. Hind wings very much reduced, cubitus absent." (Plate 21, Fig. 2.)

In plantations in north-eastern New South Wales the banana aphis is found widely distributed on both healthy and diseased plants. They are seen around the base of the pseudostem at soil level, and for several inches below the soil surface; between the sheath of the outer leaf and the pseudostem, and on young suckers just emerging from the ground. Often close examination is necessary to demonstrate their presence. Where plentiful they are also seen on the apex of the pseudostem, clustered in colonies around the heart-leaf and the bases of the petioles of the leaves. They may vary in number from a few individuals to large colonies. They usually take up a position on the most sheltered and inaccessible parts of the plant. Numerous examinations both during the winter and summer have failed to demonstrate their presence on the roots of the banana. When aphides are transferred to healthy plants, the greater number migrates to the basal region of the pseudostem. As they multiply they spread upwards towards the heartleaf. Aphides are most numerous during wet seasons; their numbers decrease rapidly during periods of drought. They flourish throughout the whole year, but the summer months are most favorable to their development. A few individuals under optimum conditions for development soon produce a dense black colony. When viewed with the unaided eye the banana aphis appears dark brown or brownish-black in colour. Both winged and wingless individuals occur in the normal colony, the wingless aphides being by far the most plentiful. The adult winged forms have the nervures delicately lined with black, which gives them a characteristic appearance.

Large quantities of "honey-dew" are produced by a colony of banana aphides. This is often seen on the leaves as fine droplets, and accumulates around the pseudostem between the sheathing bases of the leaves. In the sugary solution thus formed, yeasts develop to produce a thick slimy white paste. The large quantities of "honey dew" attract ants, which swarm over the aphid colonies. The presence of ants on a banana plant is a good indication of the association of aphides.

1. Specificity of the Banana Aphis.—Observations suggest that the banana aphis is specific in its host relationships. It is very doubtful if Pentalonia nigronervosa will live and flourish on any host other than a member of the natural order Musaceae. In heavily infested plantations the banana aphis has not been observed on any plant other than the banana. Banana aphides have never been seen on weeds or grasses growing in close proximity to stools heavily infested with aphides. Banana aphides have been transferred to the potato (Solanum tuberosum), maize (Zea mays), arrowroot (Canna edulis), canna (Canna sp.), Strelitzia sp., manila hemp (Musa textilis), and a seedbearing variety of banana (Musa banksii) indigenous to the scrubs of North Queensland, all growing under glasshouse conditions. On Musa textilis and Musa banksii the aphides flourished well. When transferred to the potato and maize they remained for only a few hours; on the arrowroot and canna the aphides lived for a week to a fortnight, but eventually either perished or deserted the plants. On Strelitzia the aphides remained for a slightly longer period, but finally disappeared. Subsequent attempts to establish colonies of Pentalonia nigronervosa on these species growing under glasshouse conditions also failed.

During March, 1926, Mr. G. H. Hardy, Walter and Eliza Hall Fellow of the Queensland University, reported having observed colonies of *Pentalonia nigronervosa* on *Strelitzia sp.* and *Ravenala sp.* growing in the Botanic Gardens, Brisbane, in the neighbourhood of banana stools. An aphis closely resembling *Pentalonia nigronervosa* was also observed on plants of *Caladium sp.* growing in a greenhouse in the same gardens. It is understood that such plants harbour aphides in abundance at least during certain times of the year, and the gardeners intimate that the aphides are of the type now recorded. It cannot yet

be definitely stated whether these aphides are to be regarded as *Pentalonia nigronervosa* or *P. caladii* V. d. Goot., the latter being characterized by having "veins not heavily bordered."

- 2. Dissemination of Aphides.—There are a number of ways by which aphides may spread through a plantation or a banana district—
  - Transfer from plant to plant in the same row or adjacent rows by the contact of foliage or the movement of aphides over the soil.
  - (2) Flight of winged individuals.
  - (3) Carriage of aphides, winged and wingless, by strong winds.
  - (4) By the transportation of aphis-infested suckers from plantation to plantation.
  - (5) By translocation of aphis-infested soil on tools and the boots of workers.
  - (6) On the clothes of workers during cultural and harvesting operations.
- 3. Role of Aphides in Transmission of Bunchy Top.—The actual part played by the banana aphis in the transmission of Bunchy Top is not definitely known. By virtue of the possession of certain characteristics or functions, it is able, after feeding on affected plants, to cause transmission of the disease, while artificial juice-transfer inoculations are unsuccessful. The relationship between the causal agent of the disease or virus, and the vector or carrier, may be purely mechanical and incidental, or the virus may pass through a definite stage of its life history within the bodies of the aphides. Two preliminary experiments have been conducted in an endeavour to learn something about this relationship.

Experiment No. 1.—A large number of banana aphides were removed with a camel-hair brush from a Bunchy Top banana plant and were transferred on to a layer of moist cotton wool in a small insect cage. After holding the aphides in this manner for 84 hours, twenty individuals were, on 5th October, 1925, transferred to each of two young healthy banana plants (free from aphides) growing in separate pots of soil under an insect cage in section B of the glasshouse. On 26th October, twenty days later, both plants were showing definite first symptoms of Bunchy Top.

It is clear from this that aphides can retain their infective capacity for some time after removal from the source of inoculum. It was not possible to keep aphides alive for a longer period under these conditions. An attempt to cultivate infective aphides on young maize plants in order to determine if the feeding on a new host would cause the aphides to become non-virulent failed on account of their inability to adapt themselves to the new host.

Experiment No. 2.—On 21st October, 1925, several thousand infective aphides were crushed up and thoroughly mixed with 30 c.cs of sterile water. The mixture was strained through muslin; the fluid was then inoculated by fine needle pricks and hypodermic syringe injections into the two youngest leaves of four young healthy banana plants growing under insect-proof glasshouse conditions. Four control plants received similar injuries, but without inoculation.

The inoculated plants were apparently unaffected by the introduction of the body juices of infective aphides. On 30th January, 1926, all the plants in the experiment were still healthy and vigorous.

The negative result of this experiment does not prove anything; but at the same time it does not strengthen the possibility of the existence of a highly infectious stage of the virus within the bodies of the aphides. The failure to secure transmission of the disease in this experiment, and also earlier by juice-transfer inoculations, is possibly due to the special requirements necessary to produce inoculation not being satisfied. The phloem region of the vascular system is probably the only region which is susceptible to infection. Brandes(7) has shown that Aphis maydis, the vector of sugar cane mosaic, in bringing about inoculation, inserts its setae into the phloem of the vascular bundles. The penetration of the tissues by the very fine setae is aided by a copious secretion from the salivary glands, and practically no structural injury results. The entry of tissues with a minimum of injury contrasts with the feeding phenomena of other insects, with coarser mouth parts, which have failed in repeated experiments to transmit the disease.

It seems reasonable that an analogous position may exist in the case of the successful inoculation of Bunchy Top by *Pentalonia nigronervosa*, in contrast to the failure to produce the disease by artificial inoculation with needle points, which, compared with aphid setae, are enormously large.

#### 11. ECOLOGY OF THE DISEASE.

Any consideration of the influence of environmental factors on the incidence of Bunchy Top should be made with three aspects in view—(a) influence of the various factors on the vector—Pentalonia nigronervosa, (b) influence on the causative agent or virus, (c) effect on the plant in its relation to the virus.

What is known concerning the climatic range of the disease does not suggest that this factor would exert any effect on its incidence or development. In Australia it has been observed under both temperate and sub-tropical conditions, while under more tropical conditions in Ceylon and Fiji it has done extensive damage. We consider that the disease would develop under all climatic conditions which will foster the growth of bananas.

Seasonal variations have a marked effect on the amount of development of Bunchy Top in a plantation in Australia. disease does make its appearance during the winter months, it is during the summer months, January to March, that the incidence of the disease is most manifest. This may be due to three factors—the more rapid development of aphides, an increase in the rate of diffusion of the virus from infected to healthy suckers in the same stool, or possibly to the warmer temperatures favouring the development of symptoms in plants which have carried infection for some time without showing signs of the disease. At any rate, it is certain that aphides are most plentiful during the warmer months of the year. The time of the year at which planting takes place in an infected area seems to have a bearing on the amount of disease which will show up during the first year. There is an earlier and more rapid development of Bunchy Top in a young plantation, when planting-up takes place in November or December, than when it is delayed till the autumn. The prevalence of aphides during the summer months is very probably the factor concerned in bringing this about.

Evidence of growers points to the fact that the rapid development of the disease is influenced by periods of rain following on a dry spell during the summer. Many growers whose plantations had been destroyed by the disease have stated that although Bunchy Top was present in their plantations, it was only under such weather conditions that the disease made rapid headway and rendered their plantations unprofitable.

Topography seems to exert an influence on the prevalence of the disease. In north-eastern New South Wales and south-eastern Queensland, bananas are grown almost exclusively on steep hillsides, on account of the occurrence of frost on the lowlands during the winter, and for the reason that it is in these regions that rich basaltic soil is available. In a plantation on a steep hillside in the early stages of the development of the disease, it is noticed that Bunchy Top is usually most plentiful in the lower, more sheltered portions of the plantation than on the exposed brow of the hill. The soil in the lower portion is usually of higher moisture content than the higher ground, the temperature is usually greater, and the growth of plants more vigorous. Aphides are usually most plentiful in such regions, the conditions being more favorable to their development than in higher exposed situations. The influence of topography on the incidence of the disease was well shown in the development of the disease in the two experimental plots at Cobaki, an account of which is given in the early part of this paper.

The fertility of the soil, in that it influences the vigour of the plant, appears to be a factor in the development of the disease. Field observations suggest that the more vigorous a plant is and the more

rapid its growth, the more susceptible it is to Bunchy Top. The disease seems to spread more rapidly through young vigorous plantations than in old worn-out plantations where growth is slow. Plantations on poorer soil do not appear to be wiped out so quickly as those on richer soil. In its earliest incidence in a large plantation where there is variation in the vigour of the different sections, Bunchy Top is usually seen to make most headway in the best part of the plantation.

The most favorable conditions for the development of Bunchy Top, therefore, appear to be during periods of rain in the summer months, on rich soil in a warm well sheltered position.

## 12. MEASURES RECOMMENDED FOR THE CONTROL OF BUNCHY TOP,

The present Investigation, in considering the matter of control, has kept in view two distinct aspects of the Bunchy Top problem, namely:—

- (a) That concerned with the conservation, as far as practicable, of the industry in the affected areas, and the problem of bringing the industry in the same areas back to its original status, as well as the resuscitation of the industry in those portions of the area where it has become moribund.
- (b) The protection of the large area in Queensland which is in no way affected with the disease.

In considering the methods of control, it is proposed to discuss them under the headings of—(1) Exclusion, (2) Protection, (3) Eradication, (4) Immunization, (5) Remedial Measures.

1. Exclusion.—The exclusion of the disease from any area can be effected, provided that none but healthy suckers are imported into that area, and that the area is sufficiently remote from any affected plantation to remove the possibility of migration or transportation of infective aphides.

The principle of exclusion should apply primarily to the unaffected areas in Queensland north of the affected zone, but should have application also to certain districts in the affected areas which occupy isolated positions and contain only a few plantations infected to a greater or less extent.

The exclusion of the disease demands, in our opinion, the efficient discharge of the following recommendations:—

(i) Prohibition of the transportation of any vegetative parts of any banana plant or of any member of the genus Musa from any part of the areas affected with Bunchy Top to the north, or to any unaffected or lightly affected area.

- (ii) Prohibition of the shifting of the vegetative parts of any species of the genus Musa from any place within a newly affected area to any other place within that area, or, otherwise prohibition of any further planting-up within the area unless with suckers obtained from a certified disease-free area.
- (iii) That no person shall be allowed to trade in suckers or to transport suckers from one area in Queensland or New South Wales to another, unless a special permit be granted by a competent Government officer, or by such persons as may be responsible for the control of Bunchy Top.
- (iv) The registration of all plantations in which any species of the genus Musa is cultivated throughout Queensland and New South Wales.
- (v) The destruction immediately of all banana plants or any member of the genus Musa in backyard or similar gardens, that is, in other than registered banana plantations, and the growing of such prohibited.
- (vi) Immediate eradication and destruction in lightly affected areas of every infected stool in which any portion or plant has shown the symptoms of the disease.
- (vii) The systematic examination by growers of all stools in plantations within a newly affected area, lightly affected area, or unaffected area.
- (viii) Prohibition of the transportation of banana fruit from any affected area to the zone north of the affected area, or out of any affected zone in which the disease may, at any time, appear, to any unaffected banana area.
  - (ix) Bunchy Top to be made a notifiable disease in all areas not so far known to be affected with the disease.
  - (x) A complete survey of the banana plantations throughout Queensland by competent inspectors; immediate attention to be given to that portion of the unaffected area which adjoins the limits of the recently discovered affected areas.
  - (xi) Illustrated lectures and practical demonstrations of an educational nature with a view to enabling banana growers to identify the disease at its earliest stages; also distribution of a clear, concise, and well illustrated pamphlet indicating the symptoms of the disease and the manner in which it can be disseminated.

We have every reason for believing, from the study of the history of the disease in Australia and the most recent spread of the disease, that the extension of Bunchy Top to new areas has been effected primarily by the importation of affected suckers, and that the part played by aphides in the transmission of the disease concerned is of importance rather in spreading the disease throughout an affected plantation, or from an affected plantation to a healthy plantation in the neighbourhood.

Efforts have been made over several years, both in Queensland and New South Wales, to check the spread of Bunchy Top by the establishment of buffer areas and the issue of regulations prohibiting the transportation of vegetative parts of the banana plant or of any species of the genus Musa from the affected zone to unaffected areas. These endeavours have so far failed to stem the spread of the disease. A study of the recent outbreak in the areas north of the Brisbane River suggests the possibility that owing to incomplete data the buffer areas and boundaries, when established, did not take in the full extent of the infected zone. The failure of the earliest attempts to confine the disease to narrow boundaries can probably be explained similarly. The need for a complete survey of plantations is thus evident. For this purpose and for other reasons, the registration of all banana plantations is essential to furnish information as to positions of plantations, many of which are isolated and inaccessible.

The presence of Bunchy Top in many backyard and similar gardens suggests that definite method of procedure is imperative in dealing with the casual cultivator of banana plants. The individual growing a few banana plants in his garden will not, generally speaking, exercise that amount of care which, on personal grounds, is necessary in the case of the individual attempting to grow bananas on a commercial basis. Also, owing to the size of the inspectorate, it would be impracticable to carry out inspections of backyard gardens in city and town areas.

The educational aspect should not be lost sight of, when it is considered that the greater proportion of growers in the unaffected areas are ignorant of or only slightly acquainted with the characteristics and early symptoms of the disease. The gazetting of the disease as notifiable in areas at present unaffected would materially assist in the ultimate control of the disease.

2. Protection.—Protection against the disease would be effective if there were available some means of keeping the plants free from aphides. Under such conditions it would be possible to preserve intact a plantation containing healthy plants, despite the proximity of plants affected with the disease.

This Investigation has given special attention to the possibilities of protection offered by the use of sprays, dusts, and various specifics.

On the experimental plots nicotine sulphate, pyrethrum powder, calcium cyanide, and chlorocide have been employed in an endeavour to protect plants against the disease. This work has been conducted in a very methodical fashion by the Horticulturist, and it may be said that the work has been done more efficiently than would be the case in any commercial undertaking, yet the results do not indicate that either spraying or dusting are efficient methods of control of the aphides. Neither do these operations produce more than partial control of the disease. Further, the cost of labour has proved prohibitive—about £34 per acre.

In view of the nature of the disease and the manner in which it is conveyed by aphides, it is clear that for any substance to be of value in the protection of plants against Bunchy Top it would be necessary for that substance to bring about a very high degree of control of the aphides, since it is their biological and not their mechanical effects that are disastrous. There are serious practical difficulties which militate against the effectiveness of spraying in combating the disease. Owing to the fact that the aphides feed on the base of the pseudostem and on young "peepers" down to several inches beneath the soil surface, and occupy most inaccessible positions between the closely adherent old leaf-sheaths of the pseudostem, it is a difficult matter to bring the spraying fluids in contact with them. Again, spraying fluids have no lasting effect, as has been demonstrated conclusively at the experimental plots. More important still is the fact that spraying becomes a really impracticable proposition in banana plantations where the rugged nature of the ground and the difficulties in the transport of water appear to be insurmountable.

We do not consider that any protective means are of any considerable importance in a serious attempt to control the spread of Bunchy Top. Such sprays as Black Leaf 40 and Derrisol may be utilized in a minor way, in supporting the scheme of eradication by killing the aphides on a diseased stool and thereby preventing their migration prior to the digging out and destruction of the stool.

3. Eradication.—Eradication of all stools affected with Bunchy Top, on a most thorough basis, appears to us to be the real solution of the problem, keeping in view the exclusion of the disease from unaffected areas, the control of the disease in lightly affected areas, and the rehabilitation of the heavily affected areas where banana growing has become perilous or moribund.

It is patent, from the mode of transmission of the disease, that eradication of all stools showing any symptoms of Bunchy Top should be an ideal towards the attainment of which every possible effort should be made if the affected areas are to again offer facilities for banana growing, and if the northern areas now free from Bunchy Top are to be conserved. The eradication of all diseased plants, in that

it removes the source of inoculum, seems to offer greater possibilities of success than any scheme which could be elaborated for the control of aphides—the carriers of the inoculum.

The disease is distinctly systemic, and we cannot over-emphasize this fact when recommending that eradication should be interpreted as meaning the removal and destruction of all parts of any stool which have developed symptoms of Bunchy Top in any of its parts. We mention this fact, since attempts are made by some growers to conserve apparently healthy plants in an affected stool by killing out only the affected parts. We are well aware that in a certain minor percentage of cases in which the killing out has happened to be done early enough, certain parts of the stool have stood up and remained healthy for a long period; that is to say, they have missed the infection from which the killed portions of the stool have suffered. The chances of success along these lines are very small, since there is ample opportunity for the spreading of the disease by aphides from the portion of the stool which first contracted the disease, independent of the possible diffusion of the virus from one member of the stool to another member of the same stool. Also from the practical stand-point it is not economical with respect to time on the part of the grower to reckon on the physiological independence of certain parts of the stool, which sometimes does obtain, and hence the impossibility of diffusion of the virus. Further, the adoption of any such procedure would so seriously damage the projected plan of eradication on a universal basis as to remove all hope of controlling the disease.

The eradication of Bunchy Top in the affected areas demands, in our opinion--

- (i) The cleaning up of all deserted plantations.
- (ii) The cleaning up of affected plantations still under cultivation or not deserted. (This includes heavily infected plantations, "still payable" plantations, and lightly infected plantations.)
- (iii) The destruction of all banana plants growing in backyard and similar gardens.
- (iv) Rigid enforcement of regulations rendering the owner of any area liable for harbouring affected stools after the lapse of a stated period from the adoption of eradication measures.
- (v) The discouragement of further planting up within the heavily affected areas for an indefinite period, that is, until such time as a clean certificate can be given for the whole of the affected area.

- (vi) That after a period of twelve months from the date of the adoption of the eradication measures, the matter of the complete destruction of all banana plants throughout the affected areas in New South Wales and Queensland, or in certain plantations bearing unfavorable reports, be considered by the body responsible for the control of Bunchy Top.
- (vii) The future replanting in the infected areas to be done only with suckers from a certified Bunchy Top-free area.

The basis on which these recommendations are made is the belief, held in the present state of our knowledge, that Bunchy Top is a specific disease confined to species of the genus Musa, that the banana aphis is the main transmitting agent, and that this aphis is confined to members of the natural order Musaceae, which, apart from the banana, is not represented in plantations in the affected area. We recognize that there are great difficulties in the way of a consummation of the scheme of eradication, chief among which is the large labour cost of removal of Bunchy Top plants over extensive areas, especially where the land is very stony and precipitous or is overgrown with weeds and lantana. The complete removal of affected stools growing on excessively stony land will demand that subsequent inspections of the land must be made, some time after the first digging out, to destroy those plants which have grown from the unremoved portion of the original stools.

In the badly infected areas of north-eastern New South Wales and south-eastern Queensland the presence of large acreages of abandoned plantations, much of which are owned by men who have no further interest in the banana industry, will call for special consideration and appropriations. Unless the administration and the work of eradication are undertaken most seriously, both by the Governments concerned and the growers, the period which must elapse before replanting can safely take place in the affected areas will be unduly prolonged, and the chances of further spread of the disease increased. For some time following the initial attempts to eradicate Bunchy Top from a district, the disease will recur, due to the overhang of infective aphides which have migrated from dug-out stools, or to the later development of the disease in stools receiving infection some time previous to the commencement of eradication. Observations suggest that in certain instances a period of dormancy may elapse before symptoms of Bunchy Top become manifest in infected stools, which would occasion for some time a persistence of the disease in a plantation which was being Nevertheless, we feel certain that if the scheme of eradication, outlined above, can be carried out as a district-measure over a period of time, the disease will gradually disappear from the area.

Attention has been given to the possibility of providing some means, other than the laborious method of digging-out, for the destruction of infected banana stools. A series of experiments, with arsenical and other poisons, has been conducted by Mr. H. Collard, Horticulturist to this Investigation, to determine whether stools could be completely killed out by this method. These experiments have demonstrated that, although poisoning is effective in destroying the above-ground parts of stools, the corms are not injured sufficiently to prevent them throwing up fresh suckers. Where individual plants in the stool are poisoned out, no effect is produced on adjoining or neighbouring suckers. The physiological independence of the various members of a stool, at least as far as the effects of poisoning are concerned, therefore defeats this method. There seems to be no other way of efficiently eradicating diseased stools than by the costly method of uprooting.

4. Immunization.—There appears to be little hope of obtaining stock satisfactorily resistant or immune to Bunchy Top.

Observations and experiments conducted thus far would indicate that all species of the genus Musa which could be grown commercially under sub-tropical conditions, including cultivated and wild species of the banana and Manilla hemp, contract the disease. Although the various species may differ slightly in respect of their susceptibility to Bunchy Top, yet this difference does not open up the possibility of being able to replace the standard Cavendish variety. The particular climatic and topographical conditions obtaining in the affected banana areas seems to militate against the successful commercial establishment of any variety other than the least tropical of all varieties—the Cavendish.

Bunchy Top, in 1891-95, devastated the plantations in Fiji, and ruined the industry. The disease still occurs there, but is by no means the serious menace that it was. At times it does happen that complete replanting becomes necessary in some plantations, owing to all the plants developing Bunchy Top. It would appear from observations made by the Horticulturist, on his visit to Fiji, that while the disease is prevalent, it affects a comparatively small percentage of plants in most plantations, although it is rampant in native gardens. It is hard to discover the cause of this, and it would be unscientific to attribute the same to actual immunity or resistance on the part of the plant. Unfortunately, the carefully selected plants which were sent from Fiji for experimental purposes were so badly affected with Beetle Borer that very little use could be made of them. The result is that the reputed resistant stock present in Fiji cannot be definitely reported on.

It is to be recognized that, despite the agnostic attitude with respect to Fijian plants, such problems as immunity and resistance

to Bunchy Top, variations with respect to the virulence of the virus, prevalence of the vector, &c., merit further consideration. Nevertheless the propagation of possibly resistant stock would require so long a period for purposes of restocking the large acreage of plantations that more immediate attention has been necessarily devoted to other methods of combating the disease.

5. Remedial Measures.—At times reports are made by growers and enterprising sellers of specifics—fortunately not so frequently now as in past years, that they have knowledge of remedies. It may be definitely stated, in the case of Bunchy Top, as in the case of other virus diseases of plants, that all reputed remedies are valueless. Once Bunchy Top has made its appearance in a plant, nothing will restore that plant to health, and immediate destruction is the only means of effecting any good as far as banana-growing is concerned.

#### 13. SUMMARY.

- 1. The Bunchy Top disease in bananas made its appearance in Australia in 1913, being introduced very probably in infected banana suckers from Fiji. It has since spread through the banana areas of north-eastern New South Wales and south-eastern Queensland, ruining the industry in many centres. All attempts made in the past to stem the march of the disease have failed. Recently Bunchy Top has appeared in isolated areas on the north coast of Queensland. The disease has also occasioned extensive damage in Fiji, Ceylon, and Egypt.
- 2. Thus far, no plant other than a member of the genus Musa has been found to be susceptible to Bunchy Top. No variety of banana is known to be highly resistant or immune to the disease. The Cavendish, the standard commercial variety in Australia, is very susceptible.
- 3. The symptoms of the disease are very characteristic; the leaves become shortened, narrow, brittle, upright, wavy, and slightly rolled and later take on a typical rosetted habit. The earliest and most definite symptom of the disease is the presence of characteristic broken dark green streaks along the secondary veins of the laminae, the midribs, and petioles. This streaking is correlated with an interesting pathological condition of the phloem region of the vascular bundles. The disease induces a disorganization of the root system of infected plants.
- 4. Microscopic examinations and isolation tests have failed to indicate the association of either bacteria, fungi, or protozoa as a causal agent. Definite evidence has been secured that Bunchy Top can be transmitted from diseased to healthy individuals by the banana

aphis Pentalonia nigronervosa Cql. Artificial juice-transfer inoculations and the contiguity of the roots and corms of diseased and healthy plants have not caused transfer of the disease. Bunchy Top is distinctly systemic in nature, and has been classified amongst the virus diseases of the "potato-leafroll" type.

- 5. The spread of the disease in north-eastern New South Wales and south-eastern Queensland has, in our opinion, been due, first to the distribution of infected suckers by planters over wide areas, and then to the dissemination of the disease to neighbouring plantations by the banana aphis.
- 6. A consideration of the possibility of controlling the disease shows that measures serving for the exclusion of the disease in unaffected areas, and from plantations in lightly affected areas, and measures for eradication of the disease from heavily and lightly affected areas, represent the only means available for controlling Bunchy Top. No protectionary measures are of value, and no immune or resistant stock is available.

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## 15. LIST OF ILLUSTRATIONS.

### Plate

- 1 Two secondary-infected plants.
- 2 Two healthy plants, same age under similar conditions.
- 3 Fig. 1. Lamina of young leaf showing along secondary veins dark green streaks.
  - Fig. 2. Lamina of similar young healthy leaf.
- 4 Fig. 1. Lamina of older Bunchy Top leaf showing advanced stages of streak.
  - Fig. 2. Lamina of similar older healthy leaf.
- 5 Fig. 1. Unfurling heart leaf of young plant showing broken whitish streaks.
  - Fig. 2. Portions of healthy and Bunchy Top leaf midribs.
- 6 Fig. 1. Young healthy plant, stage at which infective aphides transferred.
  - Fig. 2. Same plant one month later; showing first symptoms of Bunchy Top.
- 7 Fig. 1. Same plant as Plate 6, Fig. 1, about seven weeks after transfer of aphides, showing symptoms of a Bunchy Top leaf.
  - Fig. 2. Same plant two and a half months after transfer, showing several abnormal leaves.
- 8 Fig. 1. Same plant as Plate 6, Fig. 1, five months after transfer of aphides, showing rosetting of leaves.
  - Fig. 2. Stool eighteen months old, showing stunted habit and rosetting.
- 9 Fig. 1. Young primary-infected plant about four (4) months old. Fig. 2. Leaf of young primary-infected plant.
- 10 A secondary-infected plant, grown in sterilized soil under glasshouse conditions.
- 11 Fig. 1, Insect-proof glasshouse.
  - Fig. 2. Apparatus used in sterilizing soil by steam.
- Some of the plants which were infected by the aphides transferred to them five months previously.
- 13 Control plants which remained healthy.
- 14 Fig. 1. Young plant infested with aphides in colony formation.
  - Fig. 2. Young manila hemp plants, one on left showing Bunchy
    Top condition; control on right, healthy.

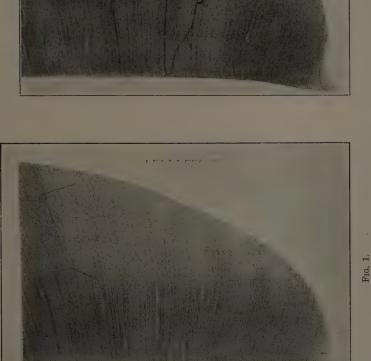
- 15 Fig. 1. Healthy (1) and Bunchy Top (3) plants which grew together without transmission of disease, owing to absence of aphides.
  - Fig. 2. Bunchy Top stool, infected at late stage, showing effect on bunch.
- View in plantation in which every stool is affected with Bunchy Top.
- 17 Fig. 1. Transverse section of vascular bundle of midrib of young healthy plant.  $\times 206$ .
  - Fig. 2. Similar section of Bunchy Top plant.
- 18 Fig. 1. Longitudinal section of phloem region of vascular bundle of midrib of young healthy plant. ×206.
  - Fig. 2. Similar section of Bunchy Top plant.
- 19 Fig. 1. Transverse section of midrib of young healthy plant, showing arrangement of vascular bundles.
  - Fig. 2. Similar section of Bunchy Top plant.
- 20 Fig. 1. Transverse section of vascular bundle of ventral periphery of midrib of leaf of young healthy banana plant.
  - Fig. 2. Similar section of Bunchy Top plant.
- 21 Fig. 1. Transverse section of phloem region of vascular bundle of interior of midrib of young Bunchy Top plant.
  - Fig. 2. Wing of banana aphis (Pentalonia nigronervosa).
- 22 Map of Queensland and New South Wales, showing infected areas.



Two secondary infected Bunchy Top plants, nine months old, which have shown the disease for four months.



Two healthy plants of the same age, grown under similar conditions.



Fro. I.—Portion of the lamina of a young leaf, photographed from the under-side in transmitted light, showing nature of the broken dark green streaks along the secondary veius, which appear as the usual first symptoms of the disease. The leaf in which these streaks first appear is otherwise normal in size, shape and behaviour. The next leaf thrown by the plant however, shows Fig 2.

Fig. 2.—Portion of the lamina of young healthy leaf photographed from the under-side in transmitted light.

marked alterations.

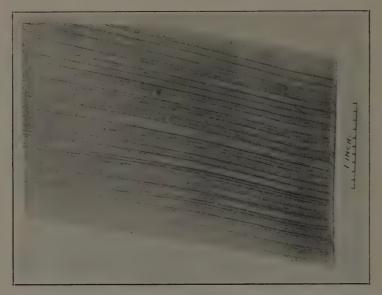
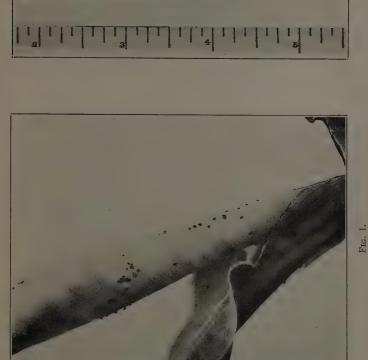


Fig. 1.—Portion of the lamina of an older Bunchy Top leaf, photographed from the under-side in transmitted light, showing advanced stage of the characteristic dark green streaking.



Fig. 2.—Portion of the lamina of an older healthy leaf photographed from the under-side in transmitted light.



Fro. 1.—Unfurling heart-leaf of young plant which is showing first symptoms of the disease as broken whitish streaks along the secondary veins of the lamina. When such a leaf has unfurled, and is viewed from the under-side with light passing through the leaf, it is observed that what were originally whitish streaks are dark green in colour.

F'IG. 2.

Fig. 2.—Portions of healthy and Bunchy Top leaf midribs. The healthy midrib is of an even pale green colour, while dark green streaking and mothling are seen in the Bunchy Top midrib in an advanced stage of the disease.



Frg. 1.—Young healthy plant, I 17 of the final transmission experiment by means of aphides, showing stage of growth at which infective aphides were transferred to the plants and also method of transfer of aphides. 29th August, 1925.



Fig. 2.—Same plant one month later. First symptoms of Bunchy Top have appeared in this plant as whitish streaks along the veins of the tightly rolled lamina of the heart-leaf. Note that the heart-leaf is about to unfurl quite normally.



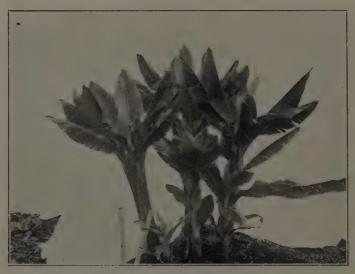
Fig. 1.—Same plant on 17th October, 1925, eighteen days after the appearance of first symptoms. The leaf marked with the arrow is the "first-symptom" leaf which is seen in the preceding figure as the heart leaf. Apart from the occurrence of broken dark green streaks along the secondary veins of the lamina, this leaf does not differ markedly from the normal preceding leaves. The following leaf, however, is showing all the symptoms of a Bunchy Top leaf. It is more narrow and upright than normal; the lamina is ridged along the main veins, is wavy and slightly rolled upwards, and showing chlorosis around the margin. Note the funnel-shaped heart leaf due to premature unfurling.



Fig. 2.—The same plant on 11th November, 1925, now showing several more abnormal leaves.



Fig. 1.—Same plant on 30th January, 1926, four months after the appearance of first symptoms, showing intensive rosetting of the leaves.



Frg. 2.—Stool eighteen months old, derived from the planting of an infected butt, showing the stunted habit and rosetting of the leaves of primary infected plants. Plants of this type never produce a bunch.



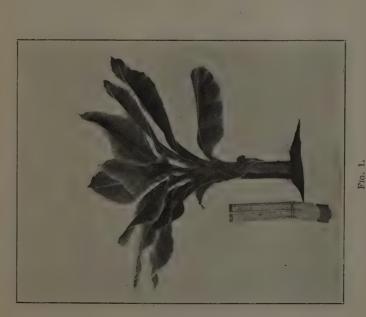


Fig. 2.

Fig. 1.—Young primary-infected Bunchy Top plant, about four months old. Note rosetting of the leaves, waviness and upward-rolling of the laminae, the ridging along the main veins and the chlorosis at the margins of the laminae.

Fig. 2.—Leaf of young primary Bunchy Top plant. Note the dark green streaking along the midrib and along the secondary veins of the lamina, the upward rolling and chlorosis of the margin of the lamina. (Photographed in transmitted and reflected light.)



PLATE 10.—A secondary-infected Bunchy Top plant, grown in sterilized soil under glasshouse conditions. This plant has shown symptoms of the disease for four months. The amount of decay present in the root system is not greater than that seen in healthy plants of the same age grown under similar conditions. The roots of this plant are entirely free from nematode galls.



Fig. 1.—Insect-proof glasshouse, in which the final transmission experiment with *Pentalonia nigornervosa*, juice-transfer inoculations, and other experiments were conducted



Fig 2.—Apparatus employed in the sterilization of soil by means of steam.



PLATE 12.—Showing some of the twenty plants which were infected with Bunchy Top by the transfer of the infective aphides, five months' previously.



PLATE 13.—Healthy control plants of the final aphis-transmission experiment planted on the same day as the plants shown in Plate 12.



Fig. 1.—Young banana plants infested with aphides (Pentalonia nigronervosa) showing colony formation. Aphides congregate on the pseudostem between the sheathing bases of the leaves and may extend on the pseudostem and young suckers into the soil for several inches.



Fig. 2.—Left.—Young Manilla hemp plant (Musa textilis), showing symptoms of Bunchy Top. Right.—Healthy control plant.





Fig. 1.—Showing one healthy and three Bunchy Top plants which have grown together in the same pot, in the absence of aphides and other insects, for over nine months without transfer of the disease.

Fig. 2.—A Bunchy Top stool showing type of bunch thrown by a plant which becomes infected at a late stage of growth: the right of the picture is seen a bunch which is borne normally.



PLATE 16.—View in a plantation in which every stool has become affected with Bunchy Top.

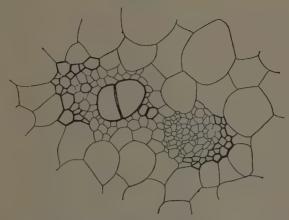


Fig. 1.—Transverse section of a vascular bundle of the midrib of a young healthy banana plant. (Drawn with the aid of the camera lucida.  $\times 206$ .)

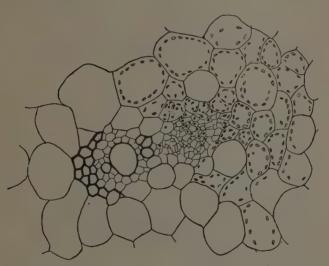
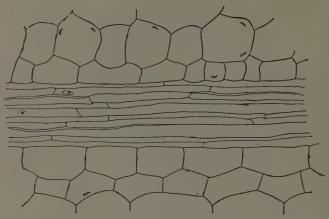


Fig. 2.—Transverse section of a vascular bundle of the [midrib of a young primary Bunchy Top plant. (Drawn with the aid of the camera lucida.  $\times 206$ .)

## NOTE.

- (1) The lack of development of fibrous tissue in the phloem region.
- (2) The presence of chlorophyll bearing cells, replacing the fibrous sheath and the abnormal development of chromatophores in the fundamental tissue at the phloem end of the vascular bundle.
- (3) The abnormally large number of nuclei present in the sieve tubes and companion cells.
- (4) Appositional growth in cells of the fundamental tissue adjacent to the phloem and the presence of chromatophores and prominent nuclei in these newly formed cells (Stained in Gentian Violet and Orange, G.)



Fro. 1.—Longitudinal section of the phloem region of a vascular bundle of the midrib of a young healthy banana plant. (Drawn with the aid of the camera lucida.  $\times 206$ .)

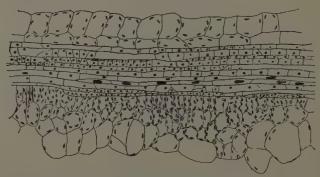


Fig. 2.—Longitudinal section of the phloem region of a vascular bundle of the midrib of a young primary Bunchy Top plant showing typical alterations induced by the disease. (Drawn with the aid of the camera lucida.  $\times 206$ .)



Fig. 1.—Photomicograph of transverse section of midrib of leaf of young healthy banana plant, showing arrangement of vascular bundles. (Hand-section, stained in dilute Gentian Violet.)



Fig. 2.—Photomicrograph of transverse section of midrib of leaf of young primary Bunchy Top plant. Note excessive development of chlorophyll tissue and alterations in region of vascular bundles. (Handsection, stained in dilute Gentian Violet.)



 $F_{\rm IG}, 1.$  —Photomicrograph of a vascular bundle of the ventral periphery of the midrib of a leaf of a young healthy banana plant.

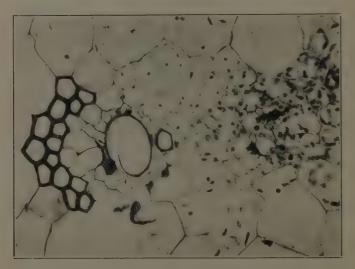


Fig. 2.—Photomicrograph of transverse section of a vascular bundle of the ventral periphery of the midrib of a leaf of a young primary Bunchy Top plant showing nature of alterations induced by the disease.

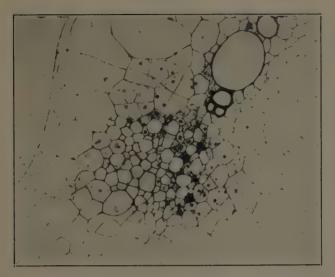


Fig. 1.—Photomicrograph of transverse section of the phloem region of a vascular bundle of the interior of the midrib of a young primary Bunchy Top plant showing appositional growth, the large number of nuclei and the absence of fibrous tissue. (Section stained in Gentian Violet and Orange G.)

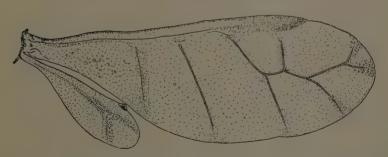


Fig. 2.—Showing wing venation of the banana aphis (Pentalonia nigronervosa). (Drawn by I. W. Helmsing.)

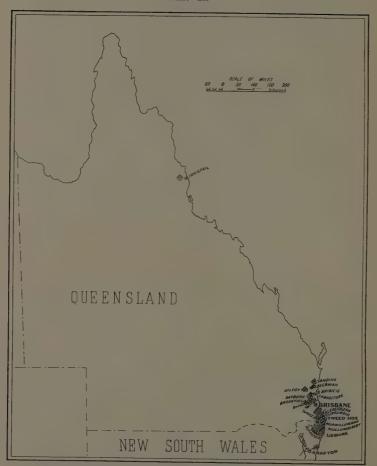
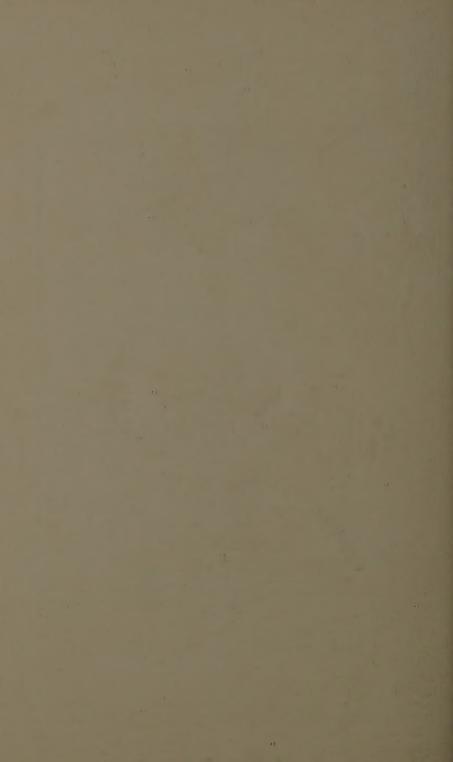


PLATE 22.-Map of Queensland and New South Wales showing infected areas.

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